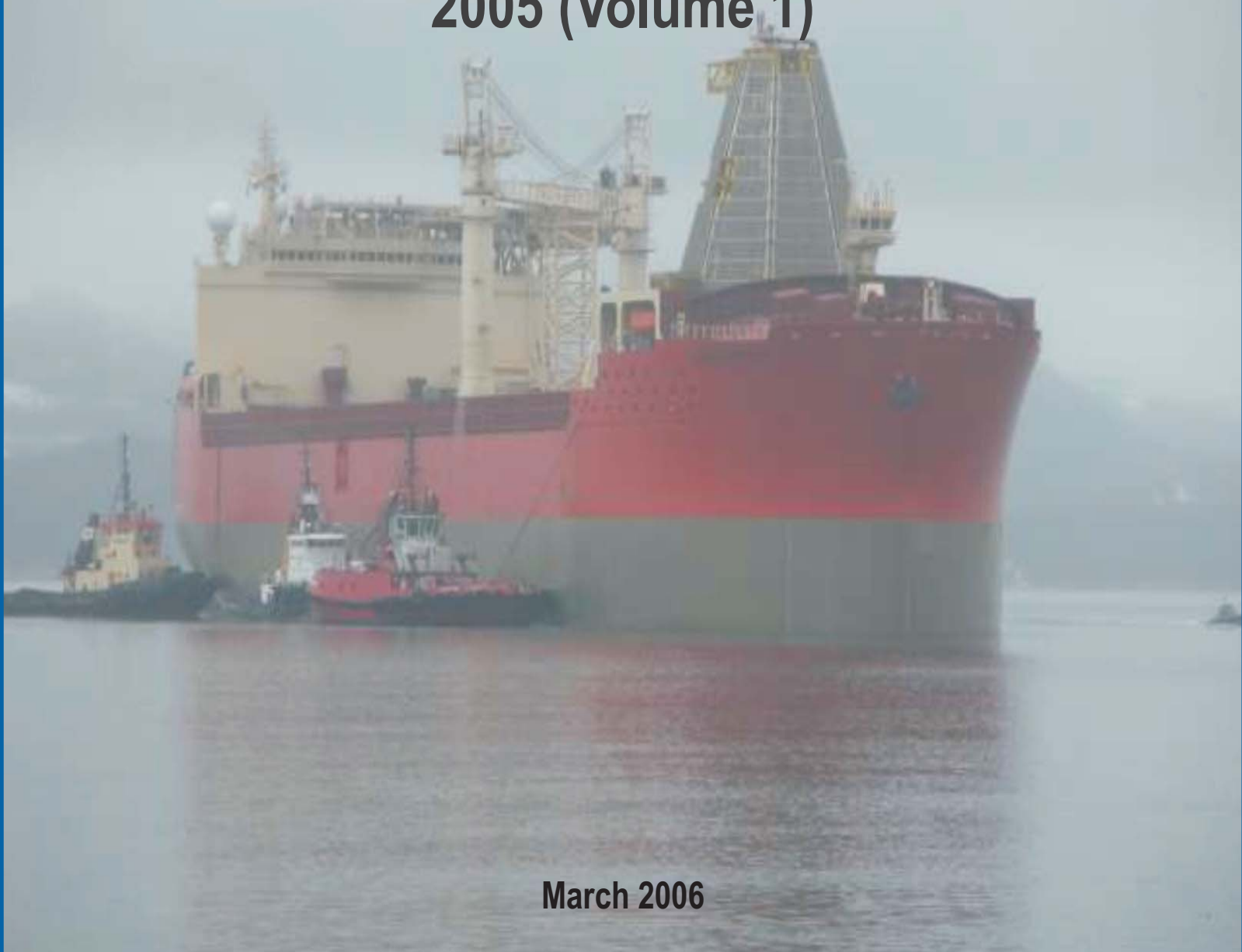




# White Rose

## Environmental Effects Monitoring Program

### 2005 (Volume 1)



March 2006

## REPORT TITLE

**2005 Environmental Effect Monitoring Program Report**

## SUBMITTED TO

**Husky Energy**  
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## Executive Summary

The White Rose Environmental Effects Monitoring (EEM) program (Husky Energy 2004) was established to fulfill a commitment made in the White Rose Environmental Impact Statement (EIS) (Husky Oil 2000). This commitment was subsequently integrated into Decision 2001.01 (C-NOPB 2001) as a condition of project approval. The design of the EEM program drew on information provided in the White Rose EIS (Husky Oil 2000), drill cuttings and produced water dispersion modeling for White Rose (Hodgins and Hodgins 2000), the White Rose Baseline Characterization program (Husky Energy 2001; 2003), stakeholder consultations and consultations with regulatory agencies. The program was designed with input from an expert advisory group that included Leslie Grattan (Environmental Planning Consultant), Dr. Roger Green (University of Western Ontario), Dr. Douglas Holdway (University of Ontario Institute of Technology), Mary Catherine O'Brien (Manager at Tors Cove Fisheries Ltd), Dr. Paul Snelgrove (Memorial University) and Dr. Len Zedel (Memorial University). The main goals of the program are to assess effects predictions made in the EIS and determine the zone of influence of project contaminants. The term "contamination" is used in this report to indicate elevated levels of a chemical as compared to background levels (GESAMP 1993).

Volumes 1 and 2 of this report provide the results of the second year of sampling for the EEM program, which was conducted in the summer of 2005. Findings are related to results of sampling conducted under the first year EEM program (Husky Energy 2005) and the Baseline Characterization program (Husky Energy 2001; 2003).

In 2005, seafloor sediments were sampled at 31 locations along transect lines radiating from the centre of the development and 13 locations surrounding the Northern, Central and Southern drill centres. Physical and chemical analyses were conducted on sediment samples. Toxicity tests that characterized whether sediments were toxic to bacteria and a marine amphipod (crustacean) species were performed. In addition, benthic invertebrate infaunal species (species living in sediment) were identified and enumerated.

Samples of a common flatfish species (American plaice) and a commercial shellfish species (snow crab) were collected in the Study Area and in four Reference Areas located approximately 28 km from the centre of the development. These samples were analyzed for chemical body burden and taste. Analyses were also performed on American plaice and snow crab Biological Characteristics (morphometric and life history characteristics), and on a variety of American plaice health indices.

Few project-related effects were noted for the 2005 EEM Program. For sediment, no project-related effects were identified for metals other than barium. However, there was clear evidence that concentrations of hydrocarbons and barium were elevated by drilling activity near drill centres. Redox levels were reduced near the Central and Southern drill centres. There was more equivocal evidence that sulphur concentrations may have been elevated near drill centres. Elevated concentrations of hydrocarbons and barium at White Rose were within the range of levels observed at other offshore oil and gas developments.

Sediment contamination did not extend beyond the zone of influence predicted by drill cuttings modeling (Hodgins and Hodgins 2000). Hydrocarbon contamination extended to between 6 and 7 km from source. Barium contamination extended to between 2 and 3 km from source. Reductions in redox levels also extended to 2 to 3 km from source. Any contamination from sulphur was limited to within 1 km from source.

Weak directional effects were noted for both hydrocarbon and barium contamination, with dispersion primarily to the southeast within 1 km from the Southern and Central drill centres. This is consistent with current records at White Rose for 2003 and 2004 (Husky Energy 2005), and with Hodgins and Hodgins (2000), who note that currents at White Rose are generally dominated by wind and tide, with a weak mean flow to the south.

There was evidence of project effects on the total abundance of *in-situ* benthic invertebrates, the relative abundance of polychaetes and the total abundance of amphipods. The spatial extent of these effects exceeded predictions made in the White Rose EIS as well as effects observed at other development sites. However, because of the power of the White Rose study design relative to other offshore EEM programs, some caution is required in inferring from this that effects on invertebrates at White Rose were worse than at other developments. The White Rose study design and data analyses were powerful enough to detect some relatively small effects. The sampling design and associated data analyses for the sediment component of the program are unique to the White Rose and Terra Nova developments, and were deliberately selected to better define the spatial extent of effects. Project effects on diversity, richness and taxa other than polychaetes and amphipods observed at other offshore developments were not detected at White Rose, but subtle depth effects and particle size (gravel content) effects on these variables were detected.

Specifically, total abundance was reduced near the Southern drill centre and decreased with increasing hydrocarbon concentrations. Decreases were more pronounced in 2005 than in 2004. Similarly, polychaete dominance was unaffected by project activity in 2004 but was reduced near the Southern drill centre in 2005. No decreases in these two variables were noted near the Central and Northern drill centres. However, relationships between the two variables and hydrocarbon concentrations in 2005 suggest that elevated hydrocarbon concentrations near the Central and Northern drill centres had some effects, even if these effects were not evident from examination spatial patterns. Relationships between total abundance and polychaete dominance versus hydrocarbon concentrations also increased in strength between 2004 and 2005.

Amphipod abundance was reduced near the Southern and Northern drill centres in 2004. Amphipod abundance was reduced near all drill centres (Southern, Northern and Central) in 2005, after drilling started at the Central drill centre. Decreases in abundance near the Northern and Southern drill centres did not intensify between 2004 and 2005. However, relationships between amphipod abundance and hydrocarbon concentrations were stronger in 2005 than 2004, suggesting at least some intensification.

Estimated zones of effects for total abundance and polychaete dominance in 2005 were between 2 and 3 km from source. These zones of effects are underestimates, since effects on both variables were observed across most of the range of detectable hydrocarbon concentrations. Total abundance decreased to approximately 65% of baseline values within the zone of effects. The relative abundance of polychaetes decreased to approximately 20% of baseline values. Effects on amphipods extended to all but the most distant stations (i.e., to 5 or more km) and across the entire range of detectable hydrocarbons. Amphipod abundance decreased to approximately 55% of baseline values within 5 km of drill centres.

For commercial fish, morphometric and life history characteristics of American plaice and snow crab collected at White Rose were similar to those of animals collected in more distant Reference Areas. Metal and hydrocarbon body burdens for both species were unaffected by project activity. Plaice and crab tissue were not tainted by sediment contamination in the Study

Area, and the general health of plaice in the Study Area, as measured through various indices, was similar to that measured in Reference Areas. Results for both plaice and crab are consistent with EIS predictions.

### **Conclusion**

Overall, project-effects at White Rose in 2005 were limited. The spatial extent and magnitude of sediment contamination were within the ranges predicted in the EIS. However, effects on benthic invertebrates were noted and these exceeded EIS predictions. Sediment contamination and effects on benthos were not coupled with effects on commercial fish. No tissue contamination was noted for crab and plaice. Neither resource was tainted, and plaice health, and plaice and crab morphometric and life history characteristics, were similar between White Rose and more distant Reference Areas.

Additional monitoring will be performed at White Rose to determine if patterns observed to date persist, intensify or moderate.

## Acknowledgements

The White Rose EEM program (2005) was led by Jacques Whitford (St. John's, Newfoundland and Labrador) under contract to Husky Energy and under the direction of Dave Taylor (Husky Energy).

Jacques Whitford led data collection, with participants including Matthew Hynest, Barry Wicks, Kathy Knox, Tim Farrell, Ian Hutton and Joseph Roberts. Fugro Jacques Geosurvey's Inc. provided geositional services for sediment collections. Benthic invertebrate sorting, identification and enumeration was led by Patricia Pocklington of Arenicola Marine (Wolfville, Nova Scotia). Chemical analyses of sediment and tissues were conducted by Maxxam Analytics (Halifax, Nova Scotia and St. John's, Newfoundland and Labrador). Particle size analysis was conducted by Jacques Whitford. Sediment toxicity was supervised by Trudy Toms of Jacques Whitford - Laboratory Division. Fish and shellfish taste tests were performed at the Marine Institute of Memorial University of Newfoundland. Fish health indicator analyses were supervised by Dr. Anne Mathieu of Oceans Ltd. (St. John's, Newfoundland and Labrador). Sediment quality, body burden and fish health data were analyzed by Dr. Michael Paine of Paine, Ledge and Associates (North Vancouver, British Columbia). Project management was executed by Dr. Elisabeth DeBlois. The Jacques Whitford analysis and reporting team included Dr. Elisabeth DeBlois, Barry Wicks, Beverley Best and Theresa Fry. Sandra Whiteway and Ellen Tracy (Jacques Whitford) and the White Rose Advisory Group reviewed the document before final printing.

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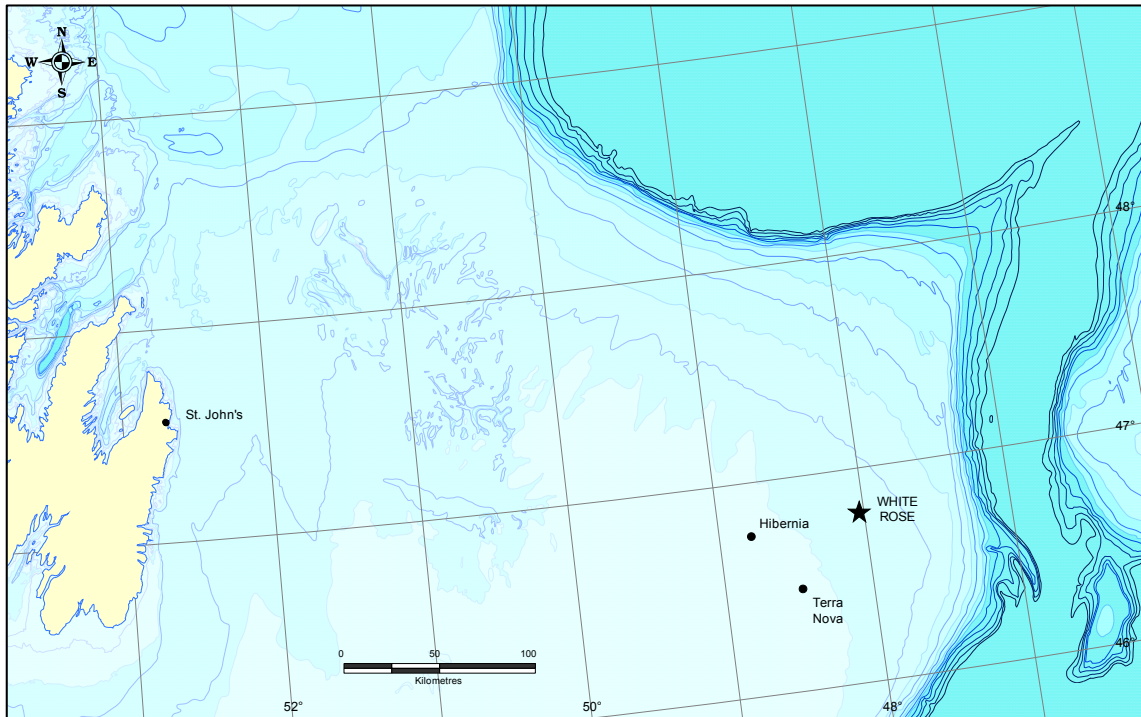
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## 1.0 Purpose

### 1.1 Project Setting and Field Layout

Husky Energy, with its joint-venture partner Petro-Canada, is developing the White Rose oilfield on the Grand Banks, offshore Newfoundland. The field is approximately 350 km east-southeast of St. John's, Newfoundland, and 50 km from both the Terra Nova and Hibernia fields (Figure 1-1). To date, development wells have been drilled at three drill centres: the Northern, Central and Southern drill centres (Figure 1-2).



**Figure 1-1 Location of the White Rose Oilfield**

### 1.2 Project Commitments

Husky Energy committed in its EIS (Part One of the White Rose Oilfield Comprehensive Study (Husky Oil 2000)) to develop and implement a comprehensive EEM program for the marine receiving environment. This commitment was integrated into Decision 2001.01 (C-NOPB 2001) as a condition of project approval.

Also, as noted in Condition 38 of Decision 2001.01 (C-NOPB 2001), Husky Energy committed, in its application to the Canada-Newfoundland and Labrador Offshore Petroleum Board (C-NLOPB), to make the results of its EEM program available to interested parties and the general public. The C-NLOPB also noted in correspondence to the White Rose Public Hearings Commissioner, Husky Energy stated its intent to make both EEM program reports and environmental compliance monitoring information “publicly available to interested stakeholders in a timely manner”.

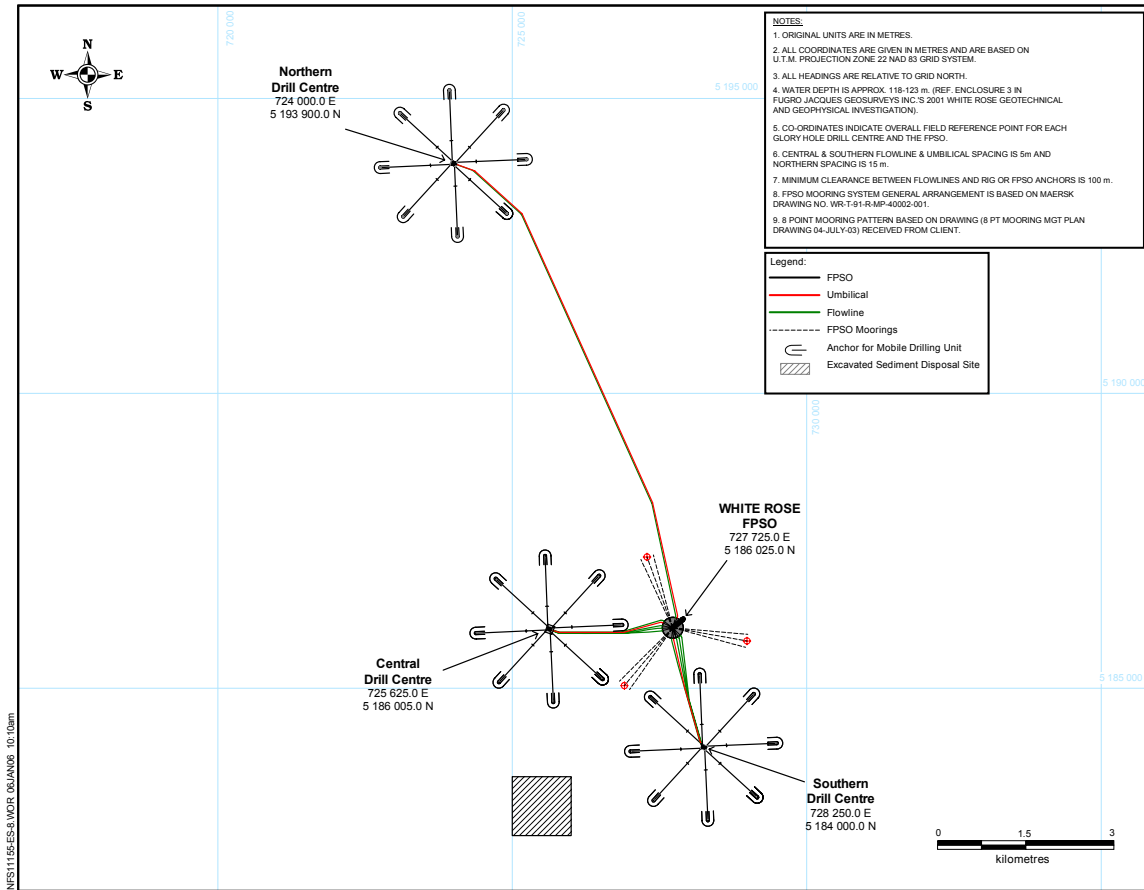


Figure 1-2 White Rose Field Layout

### 1.3 EEM Program Design

Husky Energy submitted an EEM program design to the C-NLOPB in May 2004, and this design was approved for implementation in July 2004. The design drew on information provided in the White Rose EIS (Husky Oil 2000), drill cuttings and produced water dispersion modelling for White Rose (Hodgins and Hodgins 2000), the White Rose Baseline Characterization program (Husky Energy 2001; 2003), stakeholder consultations and consultations with regulatory agencies.

The program was designed with the input an expert advisory group that included Leslie Grattan (Environmental Planning Consultant), Dr. Roger Green (University of Western Ontario), Dr. Douglas Holdway (University of Ontario Institute of Technology), Mary Catherine O'Brien (Manager at Tors Cove Fisheries Ltd.), Dr. Paul Snelgrove (Memorial University) and Dr. Len Zedel (Memorial University). The White Rose Advisory Group (WRAG) continues to provide input on interpretation of EEM results and on program refinements, as required. WRAG comments on the 2005 EEM program are provided in Appendix A.

## 1.4 EEM Program Objectives

The EEM program is intended to provide the primary means to determine and quantify project-induced change in the surrounding environment. Where such change occurs, the EEM program enables the evaluation of effects and, therefore, assists in identifying the appropriate modifications to, or mitigation of, project activities or discharges. Such operational EEM programs also provide information for the C-NLOPB to consider during its periodic reviews of the Offshore Waste Treatment Guidelines (NEB et al. 2002).

Objectives to be met by the EEM program are to:

- confirm the zone of influence of project contaminants;
- test biological effects predictions made in the EIS (Husky Oil 2000);
- provide feedback to Husky Energy for project management decisions requiring modification of operations practices where/when necessary;
- provide a scientifically-defensible synthesis, analysis and interpretation of data; and
- be cost-effective, making optimal use of personnel, technology and equipment.

## 1.5 White Rose EIS Predictions

The EIS process assesses if effects on Valued Ecosystem Components (VECs) will be significant given a series of project activities/discharges. To do so, the EIS aggregates published information on a variety of physical and biological ecosystem components. Fish and Fish Habitat, and Fisheries were identified as VECs in the White Rose EIS; and EIS predictions (Husky Oil 2000) that relate to these VECs are relevant to the White Rose EEM program. Specifically, these include predictions on physical and chemical characteristics of sediment and water, and prediction of effect on benthos, fish and fisheries (potential tainting).

In general, development operations at White Rose were expected to have the greatest effects on near-field sediment physical and chemical characteristics through release of drill cuttings, while regular operations were expected to have the greatest effect on physical and chemical characteristics of water, through release of produced water. The zone of influence<sup>1</sup> for these two waste streams was not expected to extend beyond approximately 9 km and 3 km from source for drill cuttings and produced water, respectively (Hodgins and Hodgins 2000). Effects of other waste streams (see Section 2 for details) on physical and chemical characteristics of sediment and water were considered small relative to effects of drill cuttings and produced water discharge.

Effects of drill cuttings on benthos were expected within approximately 500 m of drill centres. However, direct effects to fish populations, rather than benthos (on which some fish feed), as a result of drill cuttings discharge were expected to be unlikely. Effects resulting from contaminant uptake by individual fish (including taint) were expected to

---

<sup>1</sup> The zone of influence is defined as the zone where project-related physical and chemical alterations might occur.



range from negligible to low in magnitude and be limited to within 500 m of the point of discharge.

Effects of produced water (and other liquid waste streams) on physical and chemical characteristics of water were expected to be localized near the point of discharge. Liquid waste streams were not expected to have any effect on physical and chemical characteristics of sediment or benthos. Direct effects on adult fish were expected to be negligible.

Further details on effects and effects assessment methodologies can be obtained from the White Rose EIS (Husky Oil 2000). For the purpose of the EEM program, testable hypotheses that draw on these effects predictions are developed in Section 1.7.

## **1.6 EEM Program Components**

The two primary objectives of the White Rose EEM (Section 1.4) are to determine the zone of influence of project contaminants and test biological effects predictions made in the EIS. As such, the program will ultimately be divided into three components, dealing with effects on Sediment Quality, Water Quality and Commercial Fish species. The Water Quality component of the White Rose EEM program is currently under development (see Husky Energy 2004, 2005a) and is not dealt with in this report. Assessment of Sediment Quality includes measurement of alterations in chemical and physical characteristics, measurement of sediment toxicity and assessment of benthic community structure. These three sets of measurements are commonly known as the Sediment Quality Triad (SQT) (Chapman 1992; Chapman et al. 1987; 1991; Long and Chapman 1985). Assessment of effects on Commercial Fish species includes measurement of body burden, taint, morphometric and life history characteristics for snow crab and American plaice, and measurement of various health indices for American plaice. Components of the 2005 EEM program for White Rose are shown in Figure 1-3. Further details on the selection of monitoring variables are provided in the White Rose EEM Program Design document (Husky Energy 2004).

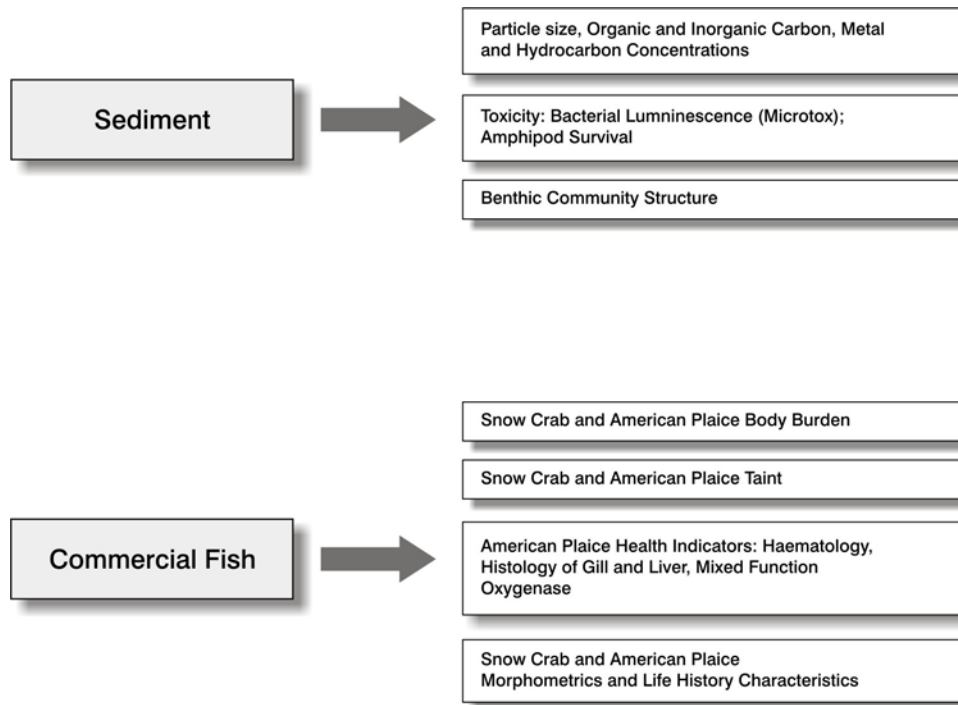


Figure 1-3 EEM Program Components

### 1.7 Monitoring Hypotheses

Monitoring, or null (H0), hypotheses have been established as part of the White Rose EEM program. Null hypotheses are an analysis and reporting construct established to assess effects predictions. Null hypotheses (H0) will always state “no effects”, even if effects have been predicted as part of the EIS. Therefore, rejection of a null hypothesis does not necessarily invalidate EIS predictions, nor should such predictions be considered a “compliance” target in this context.

The following monitoring hypotheses apply to the Sediment Quality and Commercial Fish Components of the White Rose EEM program:

- Sediment Quality:
  - H0: There will be no change in SQT variables with distance or direction from project discharge sources over time.
- Commercial Fish:
  - H0(1): Project discharges will not result in taint of snow crab and American plaice resources sampled within the White Rose Study Area, as measured using taste panels.
  - H0(2): Project discharges will not result in adverse effects to fish health within the White Rose Study Area, as measured using histopathology, haematology and mixed function oxygenase (MFO) induction.

No hypotheses were developed for American plaice and snow crab body burden, and morphometrics and life history characteristics, as these tests were considered to be supporting tests, providing information to aid in the interpretation of results of other monitoring variables (taste tests and health).

## 1.8 Sampling Design

In the Baseline Characterization (“baseline”) and the 2004 and 2005 EEM programs, sediment was sampled at discrete stations located at varying distances from drill centres, while commercial fish were sampled in the vicinity of the drill centres (Study Area) and at more distant Reference Areas (with no intermediate distances). The sediment sampling design is commonly referred to as a gradient design while the commercial fish design is a control-impact design (see Husky Energy 2004 for details).

There are some differences between the baseline, 2004 and 2005 EEM programs. A total of 48 sediment stations were sampled during baseline (Figure 1-4), 56 stations were sampled for the 2004 EEM program (Figure 1-5) and 44 stations were sampled for the 2005 EEM program (Figure 1-6); 37 stations were common to all sampling programs. As part of EEM program design (Husky Energy 2004), some redundant stations in the immediate vicinity of drill centres were eliminated for the EEM program in 2004 and 2005. These stations were sampled during baseline because the final location of drill centres had not been established. Two remote reference stations located 35 km south-southeast and 85 km northwest of White Rose were eliminated for the 2004 and 2005 EEM programs because of their distance from the development and because sediment chemistry results from baseline sampling showed that the northwest reference station might not be comparable to other stations. Two 18-km stations were eliminated because of redundancies with other stations (see Husky Energy 2004 for details).

Station additions for the 2004 and 2005 EEM programs included four reference stations at 28 km from the centre of the development, one station along the north axis at approximately 8 km from the centre of the development, three drill centre stations located approximately 300 m from each of the Northern, Central and Southern drill centres. In 2005, however, one of these stations (station S5) could not be sampled because of drilling activity at the Southern drill centre. In 2004, six drill centre stations were sampled at 1 km from the proposed location of more northerly and more southerly drill centres (see Figure 1-5). Since there are no immediate plans to drill at these centres, these stations were not sampled in the 2005 program. Table 1-1 provides a summary of changes between baseline and EEM sampling years, as well as stations name changes that were proposed in the EEM design document to simplify reporting of results.

For American plaice and snow crab, sampling for the baseline program occurred in the White Rose Study Area and in one Reference Area located 85 km Northwest of White Rose. For the EEM program, this Reference Area was replaced with four Reference Areas located roughly 28 km northwest, northeast, southwest and southeast of the development. Figures 1-7 and 1-8 provide trawl locations for the 2004 and 2005 EEM programs. The fisheries exclusion zone in 2004 was larger to accommodate possible drilling at the NN and SS drill centres. Additional information on differences between the baseline program and the EEM program can be found in the White Rose EEM design document (Husky Energy 2004)

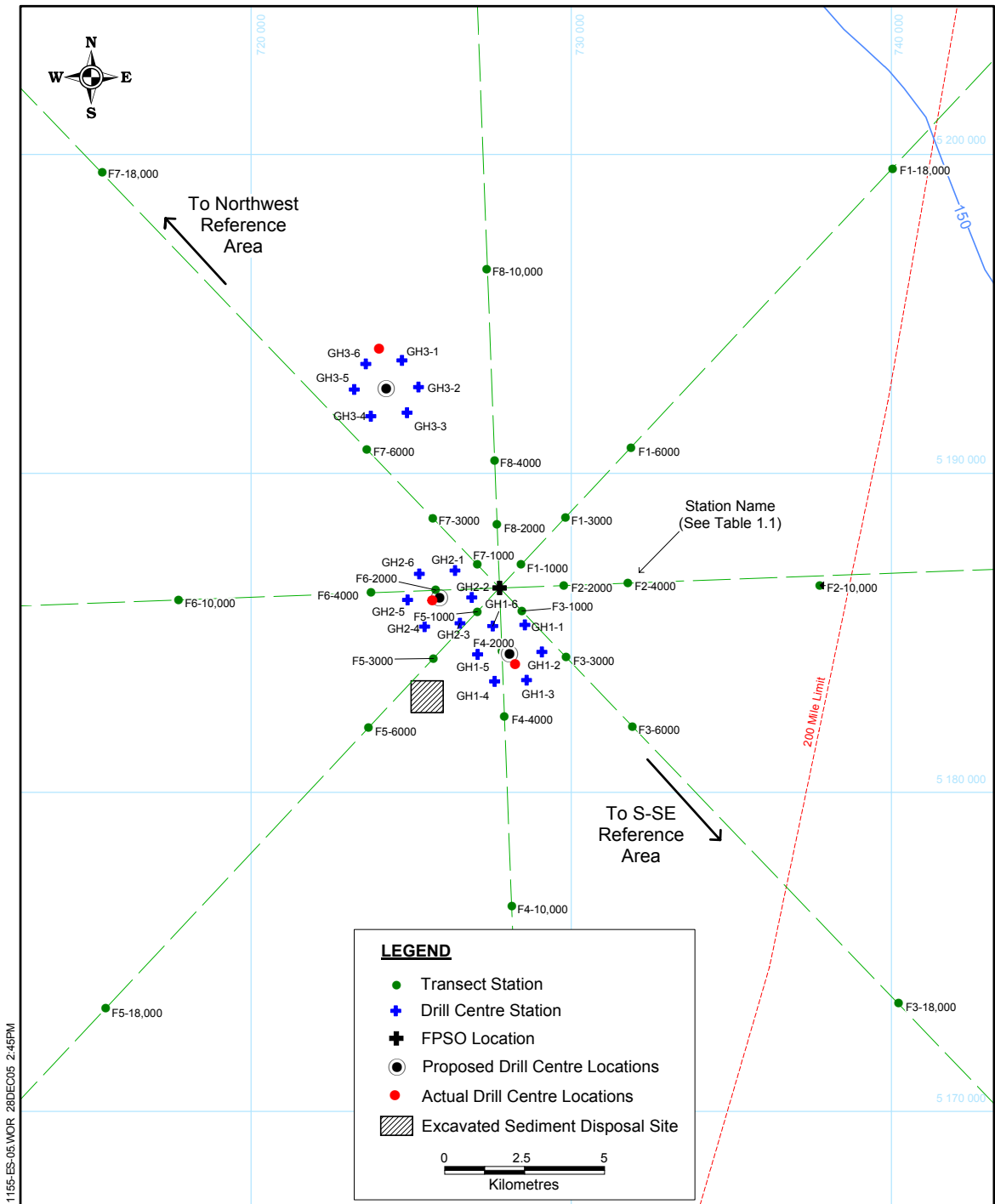


Figure 1-4 Baseline Program Sediment Stations

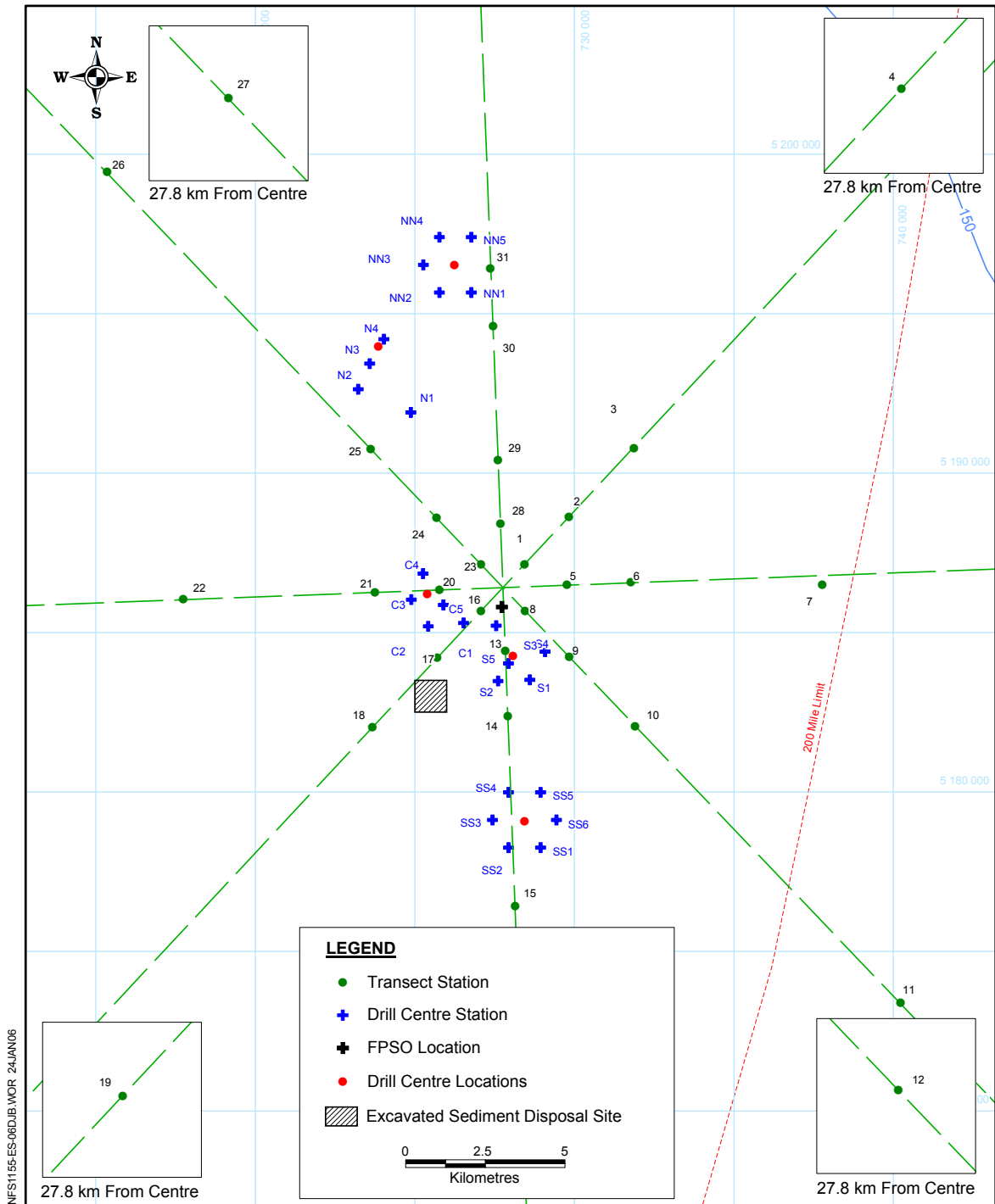
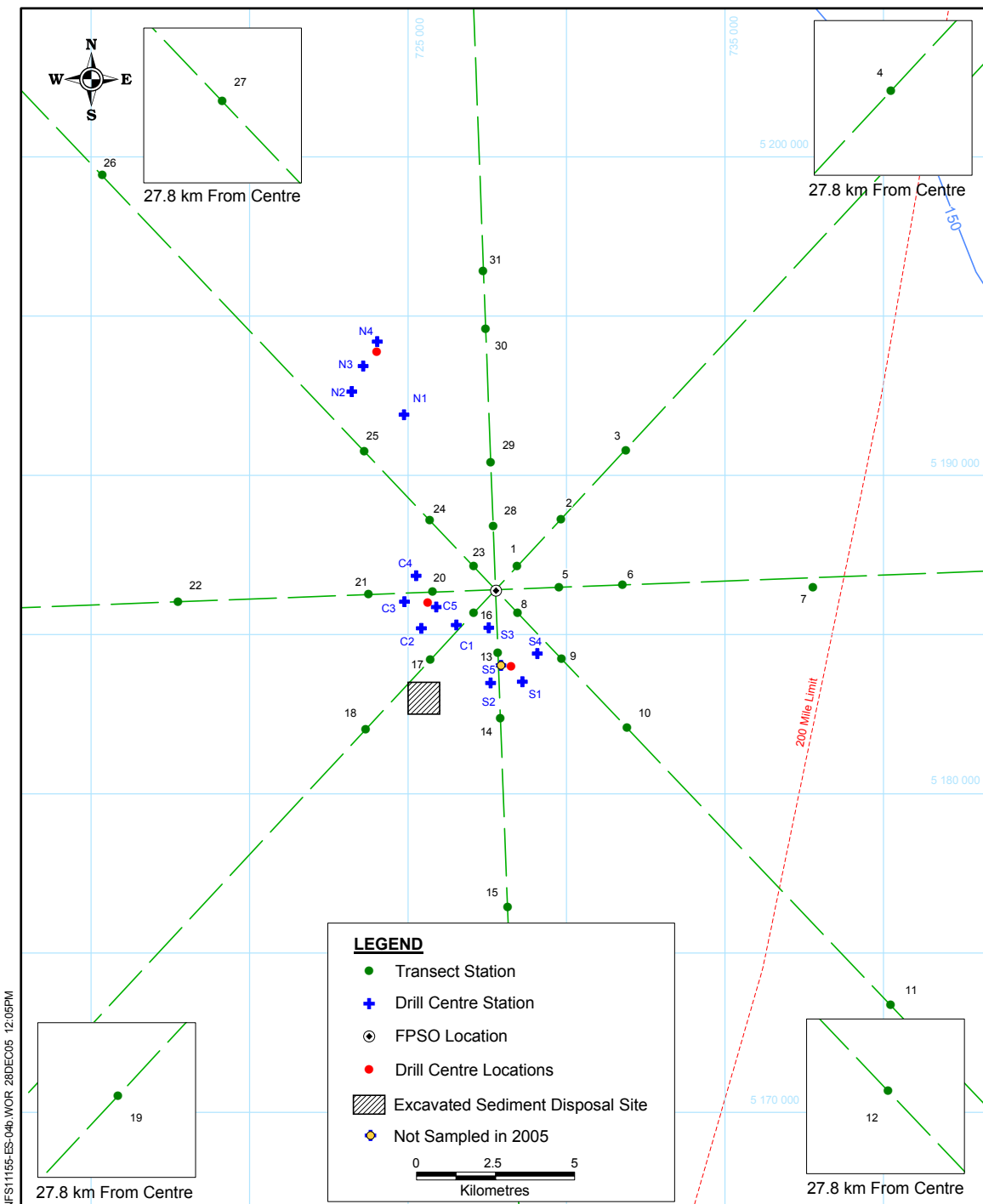


Figure 1-5 2004 EEM Program Sediment Stations



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Figure 1-6 2005 EEM Program Sediment Stations

**Table 1-1 Table of Concordance Between Baseline and EEM Stations**

EEM Station Name	Baseline Station Name	EEM Station Name	Baseline Station Name
1	F1-1,000	C1	GH2-3
2	F1-3,000	C2	GH2-4
3	F1-6,000	C3	GH2-5
4	Not Sampled	C4	GH2-6
5	F2-2,000	C5	Not Sampled
6	F2-4,000	N1	GH3-3
7	F2-10,000	N2	GH3-5
8	F3-1,000	N3	GH3-6
9	F3-3,000	N4	Not Sampled
10	F3-6,000	NN1**	Not Sampled
11	F3-18,000	NN2**	Not Sampled
12	Not Sampled	NN3**	Not Sampled
13	F4-2,000	NN4**	Not Sampled
14	F4-4,000	NN5**	Not Sampled
15	F4-10,000	NN6**	Not Sampled
16	F5-1,000	S1	GH1-3
17	F5-3,000	S2	GH1-4
18	F5-6,000	S3	GH1-6
19	Not Sampled	S4	GH1-2
20	F6-2,000	S5*	Not Sampled
21	F6-4,000	SS1**	Not Sampled
22	F6-10,000	SS2**	Not Sampled
23	F7-1,000	SS3**	Not Sampled
24	F7-3,000	SS4**	Not Sampled
25	F7-6,000	SS5**	Not Sampled
26	F7-18,000	SS6**	Not Sampled
27	Not Sampled	Deleted	GH1-1
28	F8-2,000	Deleted	GH1-5
29	F8-4,000	Deleted	GH2-1
30	F8-10,000	Deleted	GH2-2
31	Not Sampled	Deleted	GH3-1
Deleted	F1-18,000	Deleted	GH3-2
Deleted	F5-18,000	Deleted	GH3-4
Deleted	SS and NW Reference		

Notes: - \* Not sampled in 2005 because of drilling activity at the Southern drill centre  
 - \*\*Not sampled in 2005 (see text)

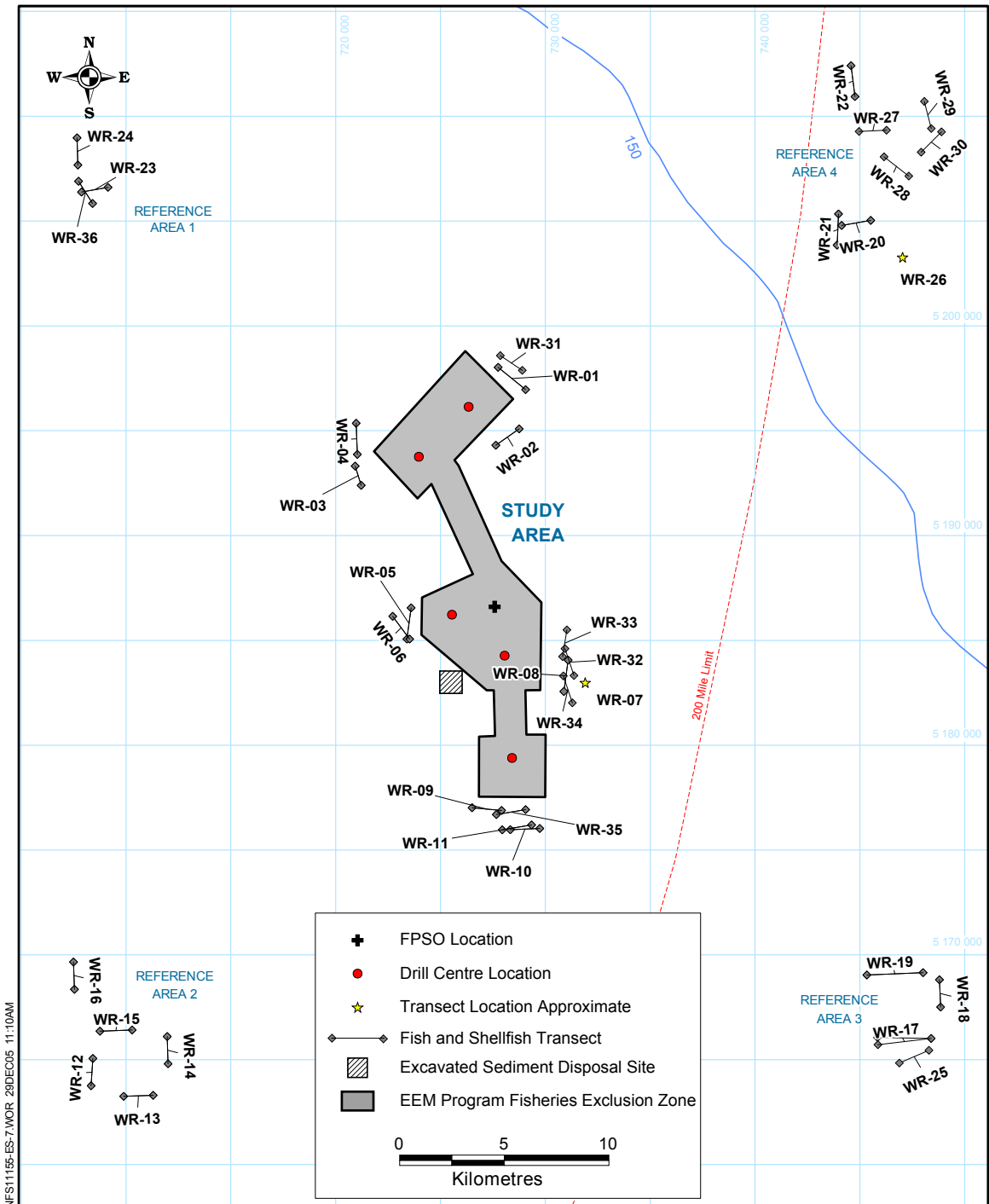


Figure 1-7 2004 EEM Program Transect Locations



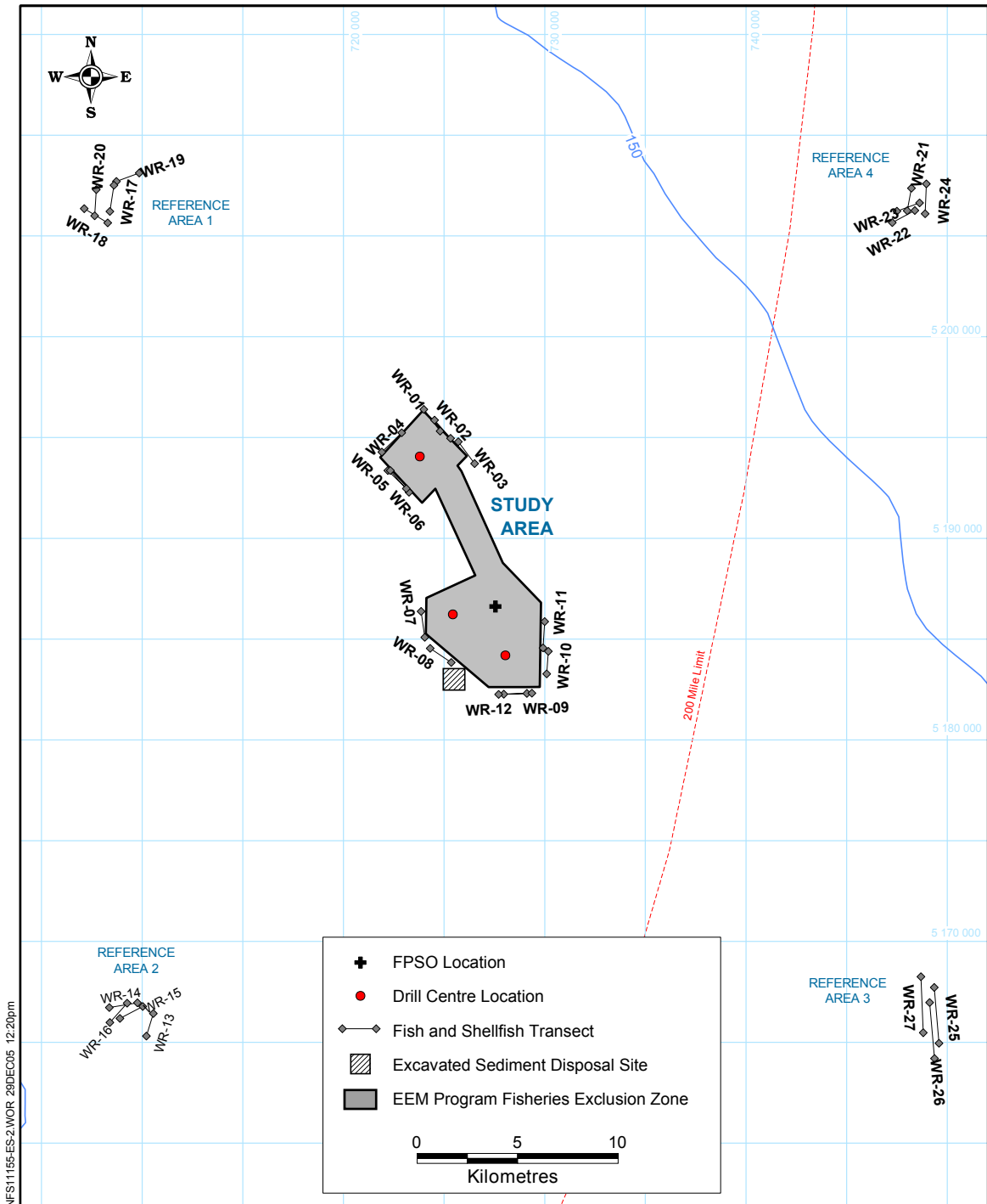


Figure 1-8 2005 EEM Program Transect Locations

## 2.0 Scope

### 2.1 Document Structure and Content

This document, *White Rose Environmental Effects Monitoring Program 2005 (Volume 1)*, provides summary results, analysis and interpretation for the White Rose 2005 EEM program. Presentation of results has been structured to provide a logical sequence of information on the physical and chemical environment, benthos and commercially important species that prey on these food sources. Where feasible, results from the baseline and the 2004 EEM programs are compared to 2005 results. A summary of findings section is included at the end of each results section. The discussion section of the report provides interpretation of results and an overall assessment of potential project effects with respect to monitoring hypotheses (Section 1.7). The discussion also includes recommendations for future EEM programs based on findings in 2005.

Most methods are provided in *Volume 1*. However, some more detailed methods as well as ancillary analyses are included in Appendices (*White Rose Environmental Effects Monitoring Program 2005 (Volume 2)*). Raw data and other information supporting *Volume 1* are also provided in *Volume 2*.

### 2.2 Background Material

The executive summary and discussion section of this document are written for a general audience. The methods and results sections assume a certain level of understanding of EEM survey design and statistical analysis. References to statistical methods used are provided in the reference section of the document (*Volume 1*). The most useful references, as well as other standard references are provided below. In addition to these, the EEM program draws on a number of general readings from the biochemical, biomedical, agriculture and hydrological literature.

Armsworthy, S.L., P.J. Cranford and K. Lee (eds.). *Offshore Oil and Gas Environmental Effects Monitoring: Approaches and Technologies*. Battelle Press, Columbus, Ohio.

*Canadian Journal of Fisheries and Aquatic Science*. 1996. Volume 53 (this volume provides reviews of GOOMEX studies).

Ellis, J.L. and D.C. Schneider. 1997. Evaluation of a gradient design for environmental impact assessment. *Env. Monitor. Assess.* 48: 157-172.

Environment Canada. 1998. *Reference Method for Determining Acute Lethality of Sediment to Marine or Estuarine Amphipods*. Report EPS 1/RM/35. Environment Canada Environmental Protection Service, Ottawa, ON.

Environment Canada. 2002. *Metal Mining Guidance Document for Aquatic Environmental Effects Monitoring*.  
<http://www.ec.gc.ca/EEM/English/MetalMining/Guidance/default.cfm>

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- van Belle, G. 2002. *Statistical Rules of Thumb*. John Wiley & Sons, New York, NY. 221 pp. (more recent rules of thumb are posted at <http://www.vanbelle.org>)

### 3.0 Definitions and Acronyms

The following acronyms are used in this report.

Acronym	Definition
ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
AR	Among Reference Areas
BC	Bray-Curtis (measure of similarity)
BS	Between Study Areas
BTEX	Benzene, Toluene, Ethylbenzene and Xylene
CCME	Canadian Council of Ministers of the Environment
CI	Confidence Interval
CL	Confidence Limit
C-NLOPB	Canada-Newfoundland and Labrador Offshore Petroleum Board
C-NOPB	Canada-Newfoundland Offshore Petroleum Board
CV	Coefficient of Variation
EBM	Exaggerated Battlement Method
EEM	Environmental Effects Monitoring
EIS	Environmental Impact Statement
EQL	Estimated Quantification Limit
EROD	7-ethoxyresorufin O-deethylase
HC	Hydrocarbon
ISQG	Interim Sediment Quality Guidelines
LOWESS	Locally Weighted Scatter-plot Smoothers
MFO	Mixed Function Oxygenase
MSDS	Material Safety Data Sheet
MDS Score	Multidimensional Score
MS(AR)	Variance Among Reference Areas
MSE	Variance Among Replicates within Areas
NMDS	Non-Metric Multidimensional Scaling
PAH	Polycyclic Aromatic Hydrocarbon
PC	Principal Component
PCA	Principal Component Analysis
PEL	Probable Effects Levels
QA/QC	Quality Assurance/Quality Control
RM	Repeated Measures
SBM	Synthetic-Based Mud
SD	Standard Deviation
SE	Standard Error
SQT	Sediment Quality Triad
SR	Study versus Reference Areas
TEL	Threshold Effects Levels
TOC	Total Organic Carbon
TPH	Total Petroleum Hydrocarbon
UCM	Unresolved Complex Mixture
VEC	Valued Ecosystem Component
WBM	Water-Based Mud
WRAG	White Rose Advisory Group

## 4.0 Project Discharges

### 4.1 Introduction

This section reports on construction, installation and drilling activities in the White Rose field. The section also summarizes the authorized discharges and spills associated with these operations.

Information on surface, mid-water and bottom currents in the White Rose area was provided in the 2004 EEM report and is not repeated here.

The purpose of this section is to provide context for the interpretation of the results from the EEM program.

### 4.2 Project Activities

Activities associated with the White Rose Development Project to date fall into four general categories:

- Construction and installation operations (completed in Fall 2005)
- Drilling operations (ongoing for the foreseeable future by one or more drill rigs)
- SeaRose Floating Production Storage and Offloading (FPSO) platform operations (ongoing for the foreseeable future)
- Supply vessel operations (ongoing for the foreseeable future)

In mid-November of 2005, producing operations (i.e., oil and gas production, storage and offloading to a tanker) began at the White Rose field once hook up, commissioning and introduction of hydrocarbons (HCs) to the FPSO SeaRose were completed.

Development drilling from the drill rig GSF Grand Banks continued in 2005 as did normal supply and standby vessel operations. Initial delineation drilling operations from the drill rig Rowan Gorilla VI took place during the months of September and October in 2005.

#### 4.2.1 Construction and Installation Operations

Construction and installation activities started in the summer of 2002 and have continued through to 2005. Activities have involved excavation of glory holes at three drill centres and subsequent installation of subsea equipment in drill centres, laying of a flow line to the Northern drill centre and installation of the spider buoy to which the FPSO was mated in late August of 2005. The remainder of the flowlines were installed, hydrostatically tested and, for the most part, dewatered in 2005 although some additional dewatering activities will occur in the future as more production and injection wells are brought on-line.

The largest physical disturbance to the seafloor to date has been the excavation of the glory holes at the three drill centres. A total of approximately 356,000 m<sup>3</sup> of seabed material, predominately sand with gravel (more than 95% sand) and some marine clays

(see Table 5.2, Section 5, for particle size diameters), was excavated and side cast within 100 m of the drill centres at the Southern and Northern drill centres. In the case of the Central drill centre, the excavated material was deposited to the seafloor between the Central drill centre and Southern drill centres (see Figure 1-2).

In 2005, installation and hook up of the SeaRose FPSO resulted in discharges of hydrostatic test and preservation fluids from subsea infrastructure and flowlines. These discharges were unavoidable, minimized to the extent possible and subject to prior regulatory approval. The components of interest in these fluids were monoethylene glycol (MEG), Tros 650, a corrosion inhibitor and a fluorescent dye.

During installation and hook up there was a maximum discharge of approximately 67 m<sup>3</sup> of MEG entrained in 273 m<sup>3</sup> of water. Smaller amounts of Tros 650 and fluorescent dye, 60 and 6 L respectively, were also discharged. In the commissioning phase, approximately 60 m<sup>3</sup> of MEG entrained in over 1000 m<sup>3</sup> of water would have been discharged along with a maximum of 600 L of Tros 650, 55 L of fluorescent dye and approximately 86 L of Transaqua HT, an ethylene glycol hydraulic fluid.

**4.2.2 Drilling Operations**

Husky Energy employs both water-based muds (WBM) and synthetic fluid-based drill muds (SBMs) in its drilling programs. WBMs are used for upper drill hole sections while SBMs are used in deeper hole sections, especially during directional drilling operations, where drilling conditions are more difficult and hole stability is critical to safety and success.

**4.2.2.1 Water-Based Drilling Discharges**

Table 4-1 summarizes the metric tonnes (MT) of drill cuttings and volumes of WBMs discharged during the conduct of development drilling by year and drill centre. The months during which drilling activities took place are also indicated.

**Table 4-1 Cumulative Cuttings and WBM Discharges from 2003 to September 2005**

Year	Drill Centre	Months with Drilling Activity												Total Cuttings Discharged (MT)	Total Muds Discharged (m <sup>3</sup> )			
		1	2	3	4	5	6	7	8	9	10	11	12					
2003	Northern																	
	Central																	
	Southern																1,037,901	1,588
2004	Northern																497,094	456
	Central																470,832	473
	Southern																1,115,359	1,784
2005	Northern																	
	Central																1,017,622	1,081
	Southern																781,281	1,380
Total Discharge at Northern Drill Centre												497,094	456					
Total Discharge at Central Drill Centre												1,488,454	1,554					
Total Discharge at Southern Drill Centre												2,934,541	4,752					
<b>Total Field Discharge</b>												<b>4,920,088</b>	<b>6,762</b>					

Note: - 2005 discharges that occurred after EEM sampling (September 2005) are not included in the above table

**4.2.2.2 Synthetic-Fluid-Based Drilling Discharges**

Table 4-2 summarizes MTs of drill cuttings and volumes of SBMs discharged during the conduct of development drilling by year and drill centre. The months during which drilling activities took place are also indicated.

**Table 4-2 Cumulative Cuttings and SBM discharges from 2003 to September 2005**

Year	Drill Centre	Months with Drilling Activity												Total Cuttings Discharged (MT)	Total Base Oil Discharged on Cuttings (m <sup>3</sup> )			
		1	2	3	4	5	6	7	8	9	10	11	12					
2003	Northern																	
	Central																	
	Southern																2,221	238
2004	Northern																473	35
	Central																1,013	117
	Southern																3,348	380
2005	Northern																	
	Central																3,514	384
	Southern																2,608	281
Total Discharge at Northern Drill Centre												473	35					
Total Discharge at Central Drill Centre												4,527	501					
Total Discharge at Southern Drill Centre												8,178	899					
<b>Total Field Discharge</b>												<b>14,213</b>	<b>1,435</b>					

Note: - 2005 discharges that occurred after EEM sampling (September 2005) are not included in the above table

**4.2.2.3 Completion Fluids**

On completion, the well bore needs to be cleaned of residual cuttings. This is done by flushing with “completion fluids” consisting of primarily sodium chloride or potassium formate brines. Table 4-3 summarizes the volumes of completion fluids discharged during the well completions by year and drill centre. The months during which these activities took place are also indicated.

**Table 4-3 Cumulative Completion Fluid discharges from 2003 to September 2005**

Year	Drill Centre	Months with Drilling Activity												Total Discharge (m <sup>3</sup> )	
		1	2	3	4	5	6	7	8	9	10	11	12		
2003	Northern														
	Central														
	Southern														
2004	Northern														
	Central														
	Southern														833
2005	Northern														
	Central														196
	Southern														1,036
Total Discharge at Northern Drill Centre													0		
Total Discharge at Central Drill Centre													196		
Total Discharge at Southern Drill Centre													1,869		
<b>Total Field Discharge</b>													<b>2,065</b>		

Note: - 2005 discharges that occurred after EEM sampling (September 2005) are not included in the above table

**4.2.2.4 Other Discharges from Drilling Operations**

From drilling operations, a total of approximately 153 m<sup>3</sup> of bilge discharges have occurred to the end of October 2005 through 15ppm rated equipment which means that there has been a maximum transfer of approximately 2.2 kg of dissolved and dispersed HCs to the ocean. Similarly, there has been approximately 3,900 m<sup>3</sup> of deck drainage reported which is managed within the 15 ppm limit specified in the Offshore Waste Treatment Guidelines (NEB 2002); meaning a transfer of a maximum of 58.6 kg of dispersed and dissolved HCs to the ocean.

Water and ethylene glycols from function testing of a seabed blowout preventer and subsea flowline valves are discharged routinely. In total, over the reporting period, approximately 265 m<sup>3</sup> of water and glycols have been discharged from these sources of which approximately 95.5 m<sup>3</sup>, or 36% of the total volume, has been the active ingredients. Note that these discharges are from the semi-submersible drill rig GSF Grandbanks that has its blow out preventer on the sea floor in contrast to a jack-up rig with its blow out preventer on the platform and hence no discharges to sea.

**4.2.3 FPSO Production Operations**

Since hookup of the SeaRose FPSO to the spider buoy at the White Rose Field, the vessel has discharged approximately 152 m<sup>3</sup> of bilge water through its 15ppm oily water treatment system to the end of October 2005. In addition, during the process of start-up, there was a total discharge of 108 m<sup>3</sup> of water from the drains system prior to its permanent connection to the slops tanks. Both these discharge volumes were managed within the 15 ppm limit of the Offshore Waste treatment Guidelines and hence the maximum transfer of dissolved and dispersed HCs to the ocean is estimated at 3.3 kg.

The majority of cooling water discharge originates with the SeaRose FPSO. This seawater is treated with chlorine to control bio-fouling and is managed such that the residual chlorine level at discharge is 0.5 ppm or less. The average concentration since



commencement of FPSO operations for the reporting period has ranged between 0.1 and 0.3 ppm. Approximately 40,320 m<sup>3</sup> of water per day is discharged from the cooling water system.

**4.2.4 Supply Vessel Operations**

All facilities and operations offshore are supported by supply and standby vessels. Normal vessel operations involve discharge of treated sewage and bilge water that contains 15 ppm or less of dissolved and dispersed oil in accordance with MARPOL (73/78) requirements. Any losses from vessel operations other than these authorized waste streams from October 2003 through October 2005 are summarized in Table 4-4.

**Table 4-4 Cumulative Losses from White Rose Offshore Operations - October 2003 through September 22, 2005**

Operation	Hydrocarbons	Drilling Fluids	Chemicals	Solid Waste	Other
<b>Drilling Operations</b>	119 L crude oil lost during well testing 85 L of hydraulic fluid lost during ROV operations	99.1 m <sup>3</sup> of SBM fluid lost	none	none	none
<b>Supply Vessel Operations</b>	35 L of hydraulic fluid lost	none	none	none	Evidence of a night collision with marine mammal? Loss of empty containers in transit to port
<b>Construction Operations</b>	67 L of hydraulic fluid lost during vessel and ROV operations	none	none	none	none
<b>FPSO Production Operations</b>	1.4 L of hydraulic fluid lost	none		none	none

Notes: - ROV = Remotely Operated Vehicle; SBM = Synthetic-Based-Mud  
 - For the purposes of this table, FPSO production operations are presumed to begin as of hook-up (August 28, 2005) whereas first oil was brought onboard in mid-November 2005

**4.2.5 Spills and Other Losses from All Sources**

Table 4-4 also summarizes the spills and other losses that have occurred during Husky operations in the White Rose Field. These losses are the result of accidental events that are reported to the C-NLOPB and are investigated to determine root causes and corrective measures.

## 5.0 Sediment Component

### 5.1 Field Collection

The sediment component of the 2005 EEM Program was conducted from September 16 to September 22, 2005, using the offshore supply vessel *Placentia*. Sampling dates for the baseline program and EEM programs are summarized in Table 5-1. Sediment stations for the baseline and EEM programs are shown in Figures 1-4 and 1-5 (Section 1). More details on the baseline survey can be found Section 1 and in Husky Energy (2001). More details on the year 1 EEM program can be found in Husky Energy (2005). Geographic coordinates and distances to drill centres for EEM stations sampled in 2005 are provided in Appendix B-1.

**Table 5-1 Date of Field Programs**

Trip	Date
Baseline Program	September 9 to September 19, 2000
EEM Program Year 1	September 26 to October 11, 2004
EEM Program Year 2	September 16 to September 22, 2005

Sediment samples were collected using a large-volume corer (mouth diameter = 35.6 cm, depth = 61) designed to mechanically take an undisturbed sediment sample over approximately 0.1 m<sup>2</sup> (0.0995 m<sup>2</sup>) of seabed (Figures 5-1 and 5-2). Three cores were performed at each station to collect sufficient sediment volume for assessment of sediment physical and chemical characteristics, toxicity and benthic community structure (SQT components; see Section 1). Sediment samples collected for physical and chemical analysis, as well as for archive, were a composite from all three cores (Figure 5-3). Sediment was sampled with a stainless steel spoon at the surface of the cores but at least 2 cm away from the corer walls (i.e. over an area of approximately 0.078 m<sup>2</sup>) and down to a depth of approximately 0.5 to 1 cm. Most of these samples were stored in pre-labelled 250-mL glass jars at -20°C. Sediment for sulphide analysis was stored at 4°C. Sediment samples collected for toxicity were taken from the top 7.5 cm of one grab and stored at 4°C, in the dark, in a 4-L pail lined with a plastic bag (amphipod toxicity) and a Whirl-Pak (bacterial luminescence). Twelve (12) of the samples that would have been used for archive were stored at 4°C, in the dark, along with toxicity samples. These samples were earmarked for chemistry analysis at the initiation and completion of each amphipod toxicity trial. Samples were obtained from stations 13, S1, S2, S4, C2, C3, C4, C5, N1, N2, N3 and N4. Sediment samples for benthic community structure analysis were collected from the top 15 cm of two grabs and stored in two separate 11-L pails<sup>2</sup>. These samples were preserved with approximately 1 L of 10% buffered formalin.

<sup>2</sup> Those chemistry samples collected from the same core as benthic community samples made up approximately 3% of the volume of sediment sampled for benthic community analysis.

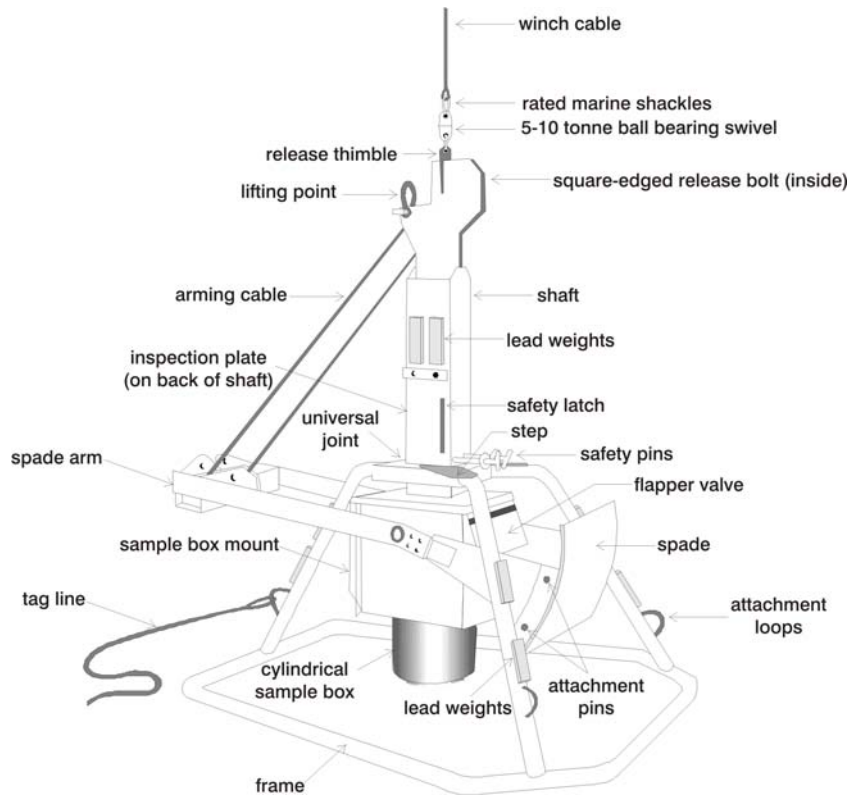
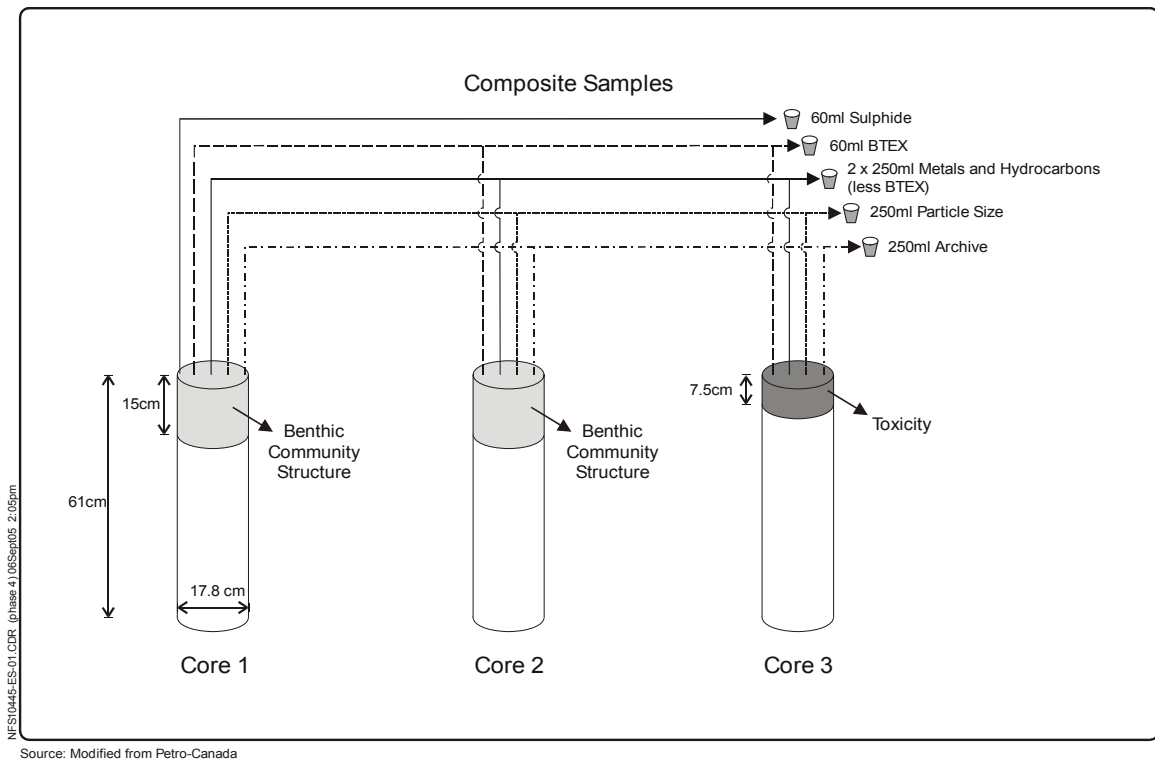


Figure 5-1 Sediment Corer Diagram



Figure 5-2 Sediment Corer



**Figure 5-3 Allocation of Samples from Cores**

Sediment chemistry field blanks composed of clean sediment obtained from Maxxam Analytics were opened at stations 9, N4 and S1. Blank vials were opened as soon the core sampler from these three stations was brought on board vessel and remained opened until chemistry samples from these stations were processed. Blank vials were then sealed and stored with other chemistry samples. Field duplicates were collected for sediment chemistry at stations 2, 15, 18, 29 and N2. Both field blanks and field duplicates were assigned randomly to stations.

The following Quality Assurance/Quality Control (QA/QC) protocols were followed for collection of samples to ensure sample integrity and prevent onboard contamination. Core samples were immediately covered with clean plastic-lined metal covers and moved to a working area near the laboratory facility. Sampling personnel were supplied with new latex gloves for each station. The laboratory facility and sampling tools were washed with isopropanol then rinsed with distilled water between each station to prevent cross-contamination between stations. Processed samples were transferred to cold storage within one hour of collection.

## 5.2 Laboratory Analysis

### 5.2.1 Physical and Chemical Characteristics

Sediment samples were processed for particle size, hydrocarbons (HCs) and metal concentration (Tables 5-2 and 5-3). Particle size analysis was conducted by Jacques Whitford in St. John's, Newfoundland and Labrador. HC and metal analyses were conducted by Maxxam Analytics in Halifax, Nova Scotia. Methods summaries from both these laboratories are provided in Appendices B-2 and B-3, respectively.

**Table 5-2 Particle Size Classification**

Size Classification (Wentworth)	Size Range (mm)	PHI Scale Range
Gravel	2 to 64	-1.000 to -6.000
Sand	0.063 to 2	3.989 to -1.000
Silt	0.002 to 0.063	8.966 to 3.989
Clay	< 0.002	< 8.986

Note: - Silt + clay fractions are referred to as "fines"

**Table 5-3 Sediment Chemistry Variables (2000, 2004 and 2005)**

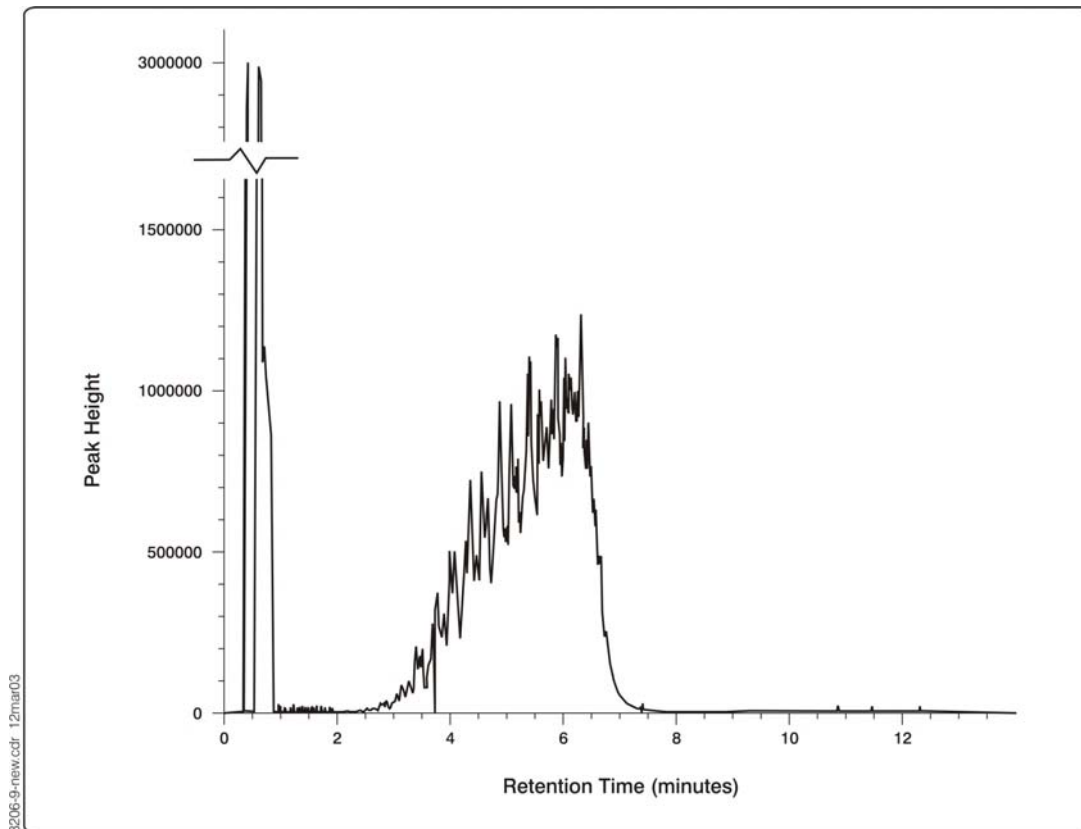
Variables	Method	2000 EQL	2004 EQL	2005 EQL	Units
<b>HCs</b>					
Benzene	Calculated	0.025	0.025	0.03	mg/kg
Toluene	Calculated	0.025	0.025	0.03	mg/kg
Ethylbenzene	Calculated	0.025	0.025	0.03	mg/kg
Xylenes	Calculated	0.05	0.05	0.05	mg/kg
C <sub>6</sub> -C <sub>10</sub>	Calculated	2.5	2.5	3	mg/kg
>C <sub>10</sub> -C <sub>21</sub> (Fuel Range)	GC/FID	0.25	0.25	0.3	mg/kg
>C <sub>21</sub> -C <sub>32</sub> (Lube Range)	GC/FID	0.25	0.25	0.3	mg/kg
<b>PAHs</b>					
1-Chloronaphthalene	GC/FID	NA	0.05	0.05	mg/kg
2-Chloronaphthalene	GC/FID	NA	0.05	0.05	mg/kg
1-Methylnaphthalene	GC/FID	0.05	0.05	0.05	mg/kg
2-Methylnaphthalene	GC/FID	0.05	0.05	0.05	mg/kg
Acenaphthene	GC/FID	0.05	0.05	0.05	mg/kg
Acenaphthylene	GC/FID	0.05	0.05	0.05	mg/kg
Anthracene	GC/FID	0.05	0.05	0.05	mg/kg
Benzo[a]anthracene	GC/FID	0.05	0.05	0.05	mg/kg
Benzo[a]pyrene	GC/FID	0.05	0.05	0.05	mg/kg
Benzo[b]fluoranthene	GC/FID	0.05	0.05	0.05	mg/kg
Benzo[ghi]perylene	GC/FID	0.05	0.05	0.05	mg/kg
Benzo[k]fluoranthene	GC/FID	0.05	0.05	0.05	mg/kg
Chrysene	GC/FID	0.05	0.05	0.05	mg/kg
Dibenz[a,h]anthracene	GC/FID	0.05	0.05	0.05	mg/kg
Fluoranthene	GC/FID	0.05	0.05	0.05	mg/kg
Fluorene	GC/FID	0.05	0.05	0.05	mg/kg
Indeno[1,2,3-cd]pyrene	GC/FID	0.05	0.05	0.05	mg/kg
Naphthalene	GC/FID	0.05	0.05	0.05	mg/kg
Perylene	GC/FID	0.05	0.05	0.05	mg/kg
Phenanthrene	GC/FID	0.05	0.05	0.05	mg/kg
Pyrene	GC/FID	0.05	0.05	0.05	mg/kg
<b>Carbon</b>					
Total Carbon	LECO	0.1	0.2	0.2	g/kg
Total Organic Carbon	LECO	0.1	0.2	0.2	g/kg
Total Inorganic Carbon	By Diff	0.2	0.3	0.2	g/kg
<b>Metals</b>					
Aluminum	ICP-MS	10	10	10	mg/kg
Antimony	ICP-MS	2	2	2	mg/kg
Arsenic	ICP-MS	2	2	2	mg/kg
Barium	ICP-MS	5	5	5	mg/kg
Beryllium	ICP-MS	5	2	2	mg/kg
Cadmium	GFAAS	0.05	0.05	0.05	mg/kg
Chromium	ICP-MS	2	2	2	mg/kg
Cobalt	ICP-MS	1	1	1	mg/kg
Copper	ICP-MS	2	2	2	mg/kg
Iron	ICP-MS	20	50	50	mg/kg
Lead	ICP-MS	0.5	0.5	0.5	mg/kg

Variables	Method	2000 EQL	2004 EQL	2005 EQL	Units
Lithium	ICP-MS	5	2	2	mg/kg
Manganese	ICP-MS	2	2	2	mg/kg
Mercury	CVAA	0.01	0.01	0.01	mg/kg
Molybdenum	ICP-MS	2	2	2	mg/kg
Nickel	ICP-MS	2	2	2	mg/kg
Selenium	ICP-MS	2	2	2	mg/kg
Strontium	ICP-MS	5	5	5	mg/kg
Thallium	ICP-MS	0.1	0.1	0.1	mg/kg
Tin	ICP-MS	2	2	2	mg/kg
Uranium	ICP-MS	0.1	0.1	0.1	mg/kg
Vanadium	ICP-MS	2	2	2	mg/kg
Zinc	ICP-MS	2	5	2	mg/kg
<b>Other</b>					
Ammonia (as N)	COBAS	NA	0.25	0.3	mg/kg
Sulphide	SM4500	NA	2	0.2	mg/kg
Sulphur	LECO	NA	0.02	0.02	%(w)
Moisture	Grav.	0.1	0.1	0.1	%

- Notes:
- The EQL is the lowest concentration that can be reliably detected within specified limits of precision and accuracy during routine laboratory operating conditions. EQLs may vary from year to year because of methods improvement and because instruments are checked for precision and accuracy every year as part of QA/QC procedures
  - NA = Not Analyzed

Within the HCs, benzene, toluene, ethylbenzene and xylenes (BTEX) are aromatic (cyclic) organic compounds, which are detected in the C<sub>6</sub>-C<sub>10</sub> range commonly referred to as the gasoline range. >C<sub>10</sub>-C<sub>21</sub> is referred to as the fuel range and is the range where lightweight fuels like diesel will be detected. The >C<sub>21</sub>-C<sub>32</sub> range is where lubricating oils (i.e., motor oil and grease), crude oil, and in some cases, bunker C oil, would be detected. HCs in all ranges include both aromatic (ring), n-alkane (straight chain) and isoalkane (branched chain) compounds. Polycyclic Aromatic Hydrocarbons (PAHs) are a diverse class of organic compounds that are composed of two or more fused aromatic benzene rings.

Gas chromatography is used to extract concentrations of HCs over the C<sub>6</sub>-C<sub>32</sub> range (see Appendix B-3). When complex HC mixtures are separated by chromatography, the more unique compounds such as the n-alkanes separate as individual peaks. Isoalkanes, on the other hand, are such a diverse group with so little difference in physical characteristics that they tend not to separate into distinct peaks in the chromatogram but rather form a "hump" in the chromatogram. This hump is often referred to as the Unresolved Complex Mixture (UCM). The drill mud base oil (PureDrill IA-35) used at White Rose is a synthetic isoalkane fluid consisting of molecules ranging from >C<sub>10</sub>-C<sub>21</sub>. Most of the components of PureDrill IA-35 form an UCM that starts around the retention time of C<sub>11</sub> n-alkane (2.25 min) and ends around the same time as C<sub>21</sub> n-alkanes (approximately 7.4 min) (Figure 5-4). The highest peaks in a chromatogram of PureDrill IA-35 have retention times similar to those of n-alkanes of C<sub>17</sub>-C<sub>18</sub> size.



**Figure 5-4 Gas Chromatogram Trace for PureDrill IA-35**

**5.2.2 Toxicity**

Jacques Whitford’s Laboratory Division in St. John’s, Newfoundland and Labrador, conducted the sediment toxicity analyses. All sediment samples were examined using the amphipod survival bioassay and the bacterial luminescence assay (Microtox). Both bioassays used whole sediment as the test matrix. Tests with lethal endpoints, in this case amphipod survival, measure survival over a defined exposure period. Tests with sublethal endpoints measure physiological functions of the test organism, such as metabolism, fertilization and growth, over a defined exposure period. Bacterial luminescence, in this case, was used as a measure of metabolism. Tests that rely on sublethal endpoints are a potential gauge of the long-term effects.

Amphipod survival tests were conducted according to Environment Canada (1998) protocols using the marine amphipod *Rhepoxynius abronius* obtained from West Beach, Whidbey Island, Washington State (USA). Tests involved five to six replicate 1-L test chambers with approximately 2 cm of sediment and approximately 800 mL of overlying water (Figure 5-5).



**Figure 5-5 Amphipod Survival Test**

Each test container was set up with 20 test organisms and maintained for 10 days under appropriate test conditions, after which survival was recorded. An additional test container was used for water quality monitoring only. Negative control sediment was tested concurrently, since negative controls provide a baseline response to which test organisms can be compared. Negative control sediment, known to support a viable population, was obtained from the collection site for the test organisms. A positive (toxic) control in aqueous solution was tested for each batch of test organisms received. Positive controls provide a measure of precision for a particular test, monitor seasonal and batch resistance to a specific toxicant, as well as standardize results to which the results for other samples may be tentatively compared. Ancillary testing of total ammonia and sulphides in overlying water was conducted by an ammonia ion selective probe and colorimetric determination, respectively.

Two sets of trials, initiated on October 25 and November 25, 2005, were required to accommodate all stations sampled at White Rose. Samples processed in the first set of trials were performed within six weeks of sample collection, meeting the requirements of sediment storage recommended by Environment Canada Guidelines (Environment



Canada 1998). Because of weather-related difficulties in obtaining amphipods for the second set of trials, holding times for samples processed during this set exceeded Environment Canada Guidelines. The 22 samples affected were those from stations: 4, 7, 8, 9, 10, 11, 12, 13, 14, 15, 18, 19, 26, 27, 30, 31, N3, N4, S1, S2, S3 and S4. For both sets of trials, three chemistry samples were sent for analysis at the initiation and completion of each trial to test if chemical characteristics of sediments were affected by storage.

The bacterial luminescence test was performed with *Vibrio fischeri*. This bacterium emits light as a result of normal metabolic activities. The Microtox assay was conducted according to the Environment Canada (2002) Reference Method EPS 1/RM/42 using the large volume solid phase assay. Analysis was conducted on a Model 500 Photometer with a computer interface. A geometric series of sediment concentrations was set up using Azur solid phase diluent. The actual number of concentrations was dependent on the degree of reduction in bioluminescence observed. Negative (clean) and positive (toxic) controls were run concurrently with the test samples. Reduction of light after 15 minutes was used to measure toxicity. Data interpretation for 2004 was conducted as outlined in Environment Canada's Reference Method (2002). Data from the 2000 (baseline) program were reexamined using the criteria outlined in Environment Canada (2002) because analyses in 2000 were conducted using earlier Environment Canada guidelines (small volume solid phase assay; Environment Canada 1992). Reinterpretation of 2000 data using Environmental Canada (2002) did not alter any of the 2000 interpretations.

All Microtox tests were initiated within six weeks of sample collection, meeting the minimal requirements of sediment storage recommended by Environment Canada Guidelines (Environment 2002).

### 5.2.2.1 Results Interpretation

The statistical endpoint for the amphipod toxicity test is the determination of whether the biological endpoint (percent survival) differs statistically from the control or reference sample, calculated using the Dunnett's Test with the TOXCALC computer program (Tidepool Scientific Software 1994). The statistical endpoint for the bacterial luminescence toxicity test is the determination of whether the biological endpoint (bioluminescence) for the sample is significantly different from the negative control (0%), calculated as the IC50<sup>3</sup> value.

Sample toxicity was assessed using standard toxicity testing statistical programs coupled with interpretation guidelines and direction provided by Environment Canada (K. Doe, pers. comm.). The amphipod survival test result for sediments were considered toxic if the endpoint (mortality) exhibited a greater than a 30% reduction in survival as compared to negative control sediment; and the result was statistically significantly different than mortality in the negative control sediment.

For the bacterial luminescence assay, as noted in above, Environment Canada has published a new reference method for Solid Phase Microtox Testing. The new reference method (Environment Canada 2002) contains new interim guidelines for assessing

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<sup>3</sup> An IC50 (50% inhibitory concentration) is the molar concentration of an agonist which produces 50% of the maximum possible inhibitory response to that agonist.

Microtox toxicity. Sediments with levels of silt/clay greater than 20% are considered to have failed this sediment toxicity test (are toxic) if the IC50 is less than 1,000 mg/L as dry solids.

For any test sediment from a particular station that is comprised of less than 20% fines and that has an IC50 (dry weight) of  $\geq 1,000$  mg/L (dry weight), the IC50 of this sediment must be compared against a sample of "clean" reference sediment or negative control sediment (artificial or natural) with a percent fines content that does not differ by more than 30% from that of the test sediment. Based on this comparison, the test sediment is judged to have failed the sediment toxicity test if, and only if, both of the following two conditions apply:

1. its IC50 is more than 50% lower than that determined for the sample reference sediment or negative control sediment; and
2. the IC50s for the test sediment and reference sediment or negative control sediment differ significantly.

There are some limitations for calculations of dry weights using the Microtox computer program (Microbics Corporation 1997) available for these tests. These limitations are both related, and unrelated, to the use of new interpretation methods for Microtox. The Microtox program does not calculate dry weights for samples that do not exhibit a reduction in bioluminescence below 197,000 mg/kg (i.e., responses  $>197,000$  mg/kg); and the program does not calculate dry weights or IC50s for samples that exhibit a dose-response relationship (hormetic response). The hormetic response (or hormesis) is a dose-response relationship in which there is a stimulatory effect at low doses and an inhibitory response at high doses resulting in a U or inverted U-Shaped dose response (Calabrese and Baldwin 2001).

Thirteen (13) White Rose samples exhibited an inverted U shaped dose response. Samples with an inverted U hormetic response showed stimulation at higher or middle dilutions followed by normal responses for lower dilutions. For these samples, dry weights could not be reported with the available software and results are reported as  $>197,000$  mg/L.

### 5.2.3 Benthic Community Structure

All 2005 samples were provided whole to Arenicola Marine Limited (Wolfville, Nova Scotia). Individual core samples were processed separately, but data were pooled for data analysis (see Section 5.3.4.2).

Sandy samples were washed through a 0.5 mm sieve. Samples with larger proportions of coarse material (gravel and shell) were elutriated and sieved by directing a high volume (1 L/s) flow of freshwater into the sample, tilting the sample bucket and catching the overflow on a 0.5 mm sieve. This washing removed the silt/clay and finer sand fractions from the samples. The procedure was adjusted to leave coarser sediment fractions in the pail. The flow suspended the less dense organisms (e.g., polychaetes) and separated small gastropods and clams which, with a suitable balance of flow in and out of the bucket, could be separated as well. Elutriation was continued until the water leaving the pail was free of organisms and when no additional heavier organisms could be seen after close examination of the sediment. Usually, larger organisms such as

scallops and propeller clams were separated manually as they were found. Barnacles and sponges were scraped off rocks. With coarser sediments such as gravels, which were occasionally encountered, a 1.2 cm mesh in combination with the 0.5 mm screen was used to aid in separating the organisms. Organisms were placed in 70% alcohol after sieving.

All samples were sorted under a stereomicroscope at 6.4x magnification, with a final scan at 16x. After sorting, substrate from 10% of samples was reexamined by a different sorter to determine sorting efficiency. Efficiency levels of 95% or better were achieved (i.e., the first sorter recovered 95% or more of the organisms recovered by both sorters combined). Wet weight biomass (g/sample) was estimated by weighing animals to the nearest milligram at the time of sorting after blotting to remove surface water. None of the samples were subsampled.

Organisms were identified to the lowest practical taxonomic level, typically to species, using conventional literature for the groups involved (Appendix B-4). All organisms were identified by Patricia Pocklington, a specialist in marine benthic invertebrate taxonomy.

Benthic invertebrate samples for 2004 were also processed by Arenicola Marine Limited. Benthic invertebrate samples from 2000 were processed by Pat Stewart of Envirosphere Ltd. Methods and the level of taxonomy were similar to those used for the 2004 and 2005 samples (see Husky Energy 2001 for details).

## **5.3 Data Analysis**

### **5.3.1 General Approach**

Analyses of sediment quality data included:

- analyses of correlations among variables for 2005 and between these variables and distances from drill centres and depth;
- comparison of distance-depth relationships among years (2000, 2004 and 2005); and
- integrated assessment of multi-year relationships between benthic invertebrate community variables and sediment physical and chemical characteristics.

The distance relationships tested “attenuation with distance” hypotheses; the integrated assessment tested “dose-response” relationships.

Given the large and complex multivariate sediment quality data set, there were reasonable alternatives available at almost every step in analysis. The general approach was to try different approaches (e.g., parametric versus non-parametric analyses; use of exposure/dose versus distance as X in regression) as opposed to minor variations of the same analysis. Suggestions from external reviews of past reports were also incorporated whenever possible. Specific analyses are described below and in Appendix B-5. For example, Appendix B-5 includes an analysis of barium concentrations normalized to aluminum concentrations specifically requested by external reviewers.

Statistical significance was defined based on the standard  $\alpha$  level or  $p \leq 0.05$ . However, emphasis was on:

- results significant at  $p \leq 0.01$  and especially  $p \leq 0.001$ ;
- strong correlations (i.e.,  $|r|$  or  $|r_s| > 0.5$  and especially  $> 0.7$ ); and
- large spatial differences, especially those attributable to potential project effects.

The White Rose program and data analyses, particularly for the multi-year data set, were powerful enough to detect some small natural and project-related effects (correlations or differences) at  $p \leq 0.05$  of lesser environmental or practical relevance. These results/effects were always reported for interested readers. However, there were many cases where strong/large natural and project-related effects were observed at low  $p$  (i.e.,  $<< 0.05$ ), and it is reasonable to place the emphasis on these less equivocal and usually more relevant results. Correlations were used as general measures of the strength of relationships (not necessarily a measure of cause-effect or environmental interest) between variables. When correlations were greater than 0.7 (or less than -0.7), parametric regressions predicting  $Y$  from  $X$  were usually provided. Any definition of “large” differences will be subjective and differ among variables. The basic approach used in this report was to ask if extreme values of some  $Y$  variable were more likely to occur at extreme values of  $X$  variables of interest (e.g., distances from drill centres; concentrations of drilling mud tracers; after versus before drilling).

All log transformations were  $\log_{10}$  rather than natural log ( $\log_e$ ) transformations. Analyses were conducted using SYSTAT 7.0.1 and Microsoft Excel 2002.

### 5.3.1.1 Analysis of 2005 Data

For analysis of 2005 data, the first step was to calculate summary statistics and any multivariate summary measures required for further analyses. Spearman non-parametric rank correlations ( $r_s$ ) within and among SQT components were then calculated and tested. Spearman  $r_s$  are parametric or Pearson correlations ( $r$ ) between the ranks of two variables. In many cases, the correlations within SQT components were tests of redundancy of variables expected to be related for statistical or natural reasons, rather than tests of meaningful environmental relationships.

Multiple regression/partial correlation analyses assessing relationships between SQT variables ( $Y$ ) and distances from drill centres and depth ( $X$ ) were then conducted. Both  $Y$  and  $X$  variables were rank-transformed (rank-rank regression or correlation). Distance measures used were distances from each drill centre (Northern, Central and Southern), and distance to the nearest drill centre (Min  $d$ ). Min  $d$  was a useful summary distance measure, particularly for plotting distance relationships in two dimensions, and often the best and simplest predictor ( $X$  variable) of  $Y$  values.

The rank-based correlation and regression analyses were useful for cases where  $Y$  values were less than EQL, and/or either  $Y$  or  $X$  values were extreme. In these cases, parametric analyses would not be appropriate without deleting some data. The rank-based analyses were able to use all the data and could be applied to almost any data set and analysis.

The rank-based analyses addressed the qualitative question: was  $Y$  more likely to increase, decrease or remain the same as  $X$  increased? In some cases, this was the only appropriate or relevant question. However, in other cases, more quantitative parametric analyses were of interest, especially for distance relationships. For these analyses, the basic model was a linear regression of  $Y$  on  $\text{Min } d (=X)$ , with  $\text{Min } d$  and often  $Y$  log-transformed. Then, two more complex models were tested:

- a “full” model with distances from each drill centre, and depth if appropriate, as  $X$ ; and
- a “hockey-stick” or threshold model with  $\text{Min } d$  as  $X$ .

Threshold distances greater than 5 and especially 10 km were difficult or impossible to estimate because most stations were within 5 km of drill centres (Appendix B-5).

Hockey-stick models assume that  $Y$  increases or decreases with increasing distance ( $X$ ) (the “shaft” of the hockey stick) up to some threshold distance ( $X_T$ ) and then does not vary with  $X$  (the “blade”). The hockey-stick regressions were useful for defining zones of influence (threshold distances or  $X_T$ ), or in some cases, indicating that zones of influence and threshold distances could not or should not be defined.

To assess the more complex models, the basic question was “did more complex models significantly reduce the residual or error variance of regression estimates relative to the simple bivariate  $Y$ - $X$  model?” (see B-5 for the test used).

The various distance models used did not directly test for directional effects. Distances from the Central and Southern drill centres were also strongly positively correlated, which made it difficult to statistically separate their effects (i.e., effects of the two drill centres were confounded to some extent). To address these issues, bubble plots (spatial distributions, with symbol sizes proportional to  $Y$  levels) were used for selected variables. Centroids (centres of  $Y$  value distributions; Section 5.3.1.2) were also calculated and compared among sample years for the same purposes.

### 5.3.1.2 Comparison Among Years

The Repeated Measures (RM) regression model described in Appendix B-5 was used to compare regressions on depth and distances from each drill centre ( $X$  variables) among years. The RM approach can only be used to analyze stations re-sampled every year. For most variables, emphasis was on the 37 stations sampled in all three years, which allowed a comparison to baseline (2000). However, some chemistry variables were not measured in 2000, so analyses were also conducted on the 42 stations sampled in both 2004 and 2005. Reference stations 4 and 19, sampled in 2004 and 2005, were excluded from the two-year RM analyses because depths at these stations were extreme, and would have a large influence on any analysis of depth effects. For the RM analyses, distances were log-transformed, some  $Y$  variables were log-transformed, and depth was not transformed.

The subsets of stations sampled in all three years, or in 2004 and 2005 when data were not available for 2000, were also used for two other purposes. Mean values and variances (Standard Deviation, SD) of  $Y$  were plotted against year to qualitatively assess net changes over the entire sample set over time. Centroids for  $Y$  variables were

calculated for each year. Appendix B-5 provides detailed methods for calculation of centroids, which removed any effects of natural/methodological differences over time occurring at all or most stations. Basic questions were:

1. Where was the location of the “average Y value” (Y centroid) relative to the location of the “average station” (sampling centroid)?
2. Did Y centroid locations change in response to onset of drilling at the Northern and Southern drill centres prior to 2004, and onset of drilling at the Central drill centre between 2004 and 2005 sampling?

Centroids were particularly useful for assessing cumulative drilling effects from all active centres, and directional and other spatial effects unrelated to distance or drilling activity.

There was evidence from the RM regressions that relationships between some Y variables and distances from active drill centres changed in strength from 2004 to 2005 (i.e., effects increased or decreased). Therefore, for selected Y variables, threshold distances ( $X_7$ ) were compared between 2004 and 2005, to qualitatively assess if the zone of influence had changed. These analyses included all stations sampled within each year. The distance measure used was distance from the nearest active drill centre (Northern and Southern in 2004; Northern, Central and Southern in 2005).

### 5.3.2 Physical and Chemical Characteristics

#### 5.3.2.1 Groups of Variables

Physical and chemical sediment characteristics were divided into four groups of related variables:

- sediment particle size and total organic carbon (TOC) content;
- known drilling mud tracers and constituents (barium,  $>C_{10}-C_{21}$  HCs, and possibly  $>C_{21}-C_{32}$  HCs);
- metals other than barium; and
- other variables (ammonia, sulphides, sulphur, redox).

Sediment particle size was expressed as % contributions of gravel, sand and fines (silt + clay). Both fines and TOC content could be altered by drilling activity. Water-based drilling muds (WBMs) and synthetic-based muds (SBMs) are finer than the predominantly sand substrate on the Grand Banks, and SBMs have a higher organic carbon content than natural substrates. Particle size, as a physical habitat variable, and TOC, as an indicator of food availability for deposit and filter feeders, can also affect benthic invertebrate communities.

Barium, as barium sulphate (barite), is a constituent of both WBMs and SBMs. SBMs have elevated concentrations of  $>C_{10}-C_{21}$  HCs, which rarely or never occur at detectable levels in natural sediments on the Grand Banks.  $>C_{21}-C_{32}$  HCs are not a major constituent of SBMs but could originate from other anthropogenic sources (e.g., deck discharges). However, when  $>C_{10}-C_{21}$  HC concentrations are high, there is also an

analytical “spill-over” effect, with some  $>C_{10-C_{21}}$  HCs appearing as  $>C_{21-C_{32}}$  HCs in chromatographs.

Metals other than barium, particularly aluminum (which occurs naturally at high concentrations in marine sediments), were primarily treated as “reference” metals, or indicators of the natural variance of barium concentrations that might be expected in the absence of drilling.

Sulphur, as sulphate in barite, is an important constituent of drilling muds, but also occurs naturally at high levels. Ammonia and sulphide levels are typically high, and redox levels low, in sediments where decomposition or degradation of natural or synthetic organic matter is extensive. High ammonia, sulphur and sulphide levels, and low redox levels, can adversely affect toxicity test organisms and *in situ* invertebrate communities.

### 5.3.2.2 Statistical Analysis

For analysis of 2005 data, Spearman rank correlations ( $r_s$ ) were calculated within and among groups of sediment physical and chemical variables. Rank-rank distance-depth regressions were also tested. In these analyses, Y values less than EQL were treated as tied for the lowest rank. Parametric distance-depth and hockey-stick models were also tested for the two tracers (barium and  $>C_{10-C_{21}}$  HCs) and redox, which were strongly affected by distance from the drill centres.

Barium, fines, TOC, metals other than barium (i.e., Metals PC1 scores; see below), and aluminum (Y) values were compared among years in RM regression models based on the 37 stations sampled in all three years (2000, 2004, 2005). These Y variables, plus ammonia, sulphur (not measured in 2000) and  $>C_{10-C_{21}}$  HCs (not detected in 2000), were also analyzed in RM regression models based on the 42 stations sampled in both 2004 and 2005 (stations 4 and 19 excluded). All Y variables except Metals Principal Component 1 (PC1) were log-transformed. Estimates of the zones of influence (threshold distances:  $X_7$ ) for the two tracers (barium,  $>C_{10-C_{21}}$  HCs) were also compared between these two years.

Principal Components Analysis (PCA<sup>4</sup>) was used to derive a summary measure of concentrations of metals other than barium for analyses of 2005 and multi-year data. Metals analyzed were aluminum, chromium, iron, lead, manganese, strontium, uranium, vanadium and zinc. Except for zinc, these metals were detected in every sample in all three sample years. Zinc was detected in every sample in 2000 and 2005, when EQLs were 2 mg/kg, but was not detected in 10 (of 56) samples from 2004 when EQLs were 5

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<sup>4</sup> PCA identifies the major axis of covariance (PC1) among the original variables (i.e., concentrations of the nine metals), which is also the major axis of variance among samples (i.e., stations). The minor axis (PC2) is the axis accounting for the largest amount of remaining covariance among variables and variance among samples that is independent of (uncorrelated with) PC1. Positions of samples along the PC axes can be expressed as scores (weighted averages of original variable values), and the scores used for further analyses. The scores are standardized, so that the overall mean is 0 with SD = 1. Metal concentrations were log-transformed prior to conducting the PCA. The sediment metal and other PCAs in this report were based on correlation rather than covariance matrices.

mg/kg. The 2004 concentrations less than EQL were set at  $\frac{1}{2}$  EQL, which introduced an analytical or artificial source of variance for concentrations less than 5 mg/kg. This variance should be relatively trivial, given that eight other metals were included in the PCA and 124 of 146 zinc concentrations were greater than or equal to 5 mg/kg.

In 2000 and 2004, EQLs for  $>C_{10}-C_{21}$  HCs were reported as 0.25 mg/kg. In 2005, EQLs were reported as 0.3 mg/kg. The change in EQL was simply rounding to better reflect the precision of the measurements; the analytical method did not change. For statistical analyses, all concentrations less than the 2005 EQL of 0.3 mg/kg were set at  $\frac{1}{2}$  that EQL or 0.15 mg/kg.

Two sulphur concentrations less than EQL of 0.02%, one in 2004 and one in 2005, were set at EQL. These values were not set at  $\frac{1}{2}$  EQL because the two-fold difference between  $\frac{1}{2}$  EQL and EQL would be as large as differences among other values (most 0.02 to 0.04%).

### 5.3.3 Toxicity

No analyses of results for 2004 and 2005 bacterial toxicity tests were conducted because all samples were non-toxic with  $IC_{50} > 197,000$  mg/kg (the highest concentration tested).

In 2005, there was one sediment sample toxic to amphipods, and one other sample with low survival (67.5% versus more than 80% for all other 2005 samples). Rank correlations between amphipod survival in toxicity tests, distances from the drill centres and sediment physical and chemical characteristics were calculated. Characteristics of the two stations with low amphipod survival were also compared with overall medians to determine if the two samples were “unusual” in some respect(s).

### 5.3.4 Benthic Community Structure

#### 5.3.4.1 Groups of Variables

Benthic community variables analyzed were:

- total abundance and standing crop (wet weight of all invertebrates recovered);
- taxonomic richness, diversity and evenness;
- multivariate community composition measures (see Section 5.3.4.2); and
- absolute abundance (i.e., numbers) of amphipods.

Absolute abundance of amphipods was treated as a separate variable because these sensitive organisms were relatively rare. Consequently, variance in amphipod numbers had a limited effect on overall community composition. In 2004, relative (%) amphipod abundances were analyzed and appeared to be affected by drilling (Husky Energy 2005). However, these effects may have been under- or overestimated, because natural or project-related changes in abundances of more common taxa will strongly affect amphipod abundances as a % of total abundance.



Nemertean, nematodes, oligochaetes, ostracods and copepods were excluded from all variables except standing crop. These small organisms are poorly recovered with the 0.5 mm mesh used. Most of the excluded organisms would have made a negligible contribution to standing crop because of their small size<sup>5</sup>.

### 5.3.4.2 Statistical Analysis

#### Preliminary Analysis

For all analyses of invertebrate communities, abundances of each taxon in the two cores collected at each station were summed (i.e., variable values were “per station” rather than “per sample”). Genera and species within families (or occasionally higher taxonomic levels) were pooled and families used as the basic taxonomic unit for analyses. For the White Rose samples, there was good agreement at the family level between the taxonomist used in 2000 and the taxonomist used in 2004 and 2005. At lower taxonomic levels (i.e., genus and species), there were some differences, mostly attributable to differences in the taxonomic level the two taxonomists were willing or able to use, especially for juveniles, and the treatment of uncertain identifications. Appendix B-4 provides abundances of lower-level taxa (usually species) for the 2005 samples. Family assignments of lower-level taxa were standardized by first using families from Gosner (1971), a general East coast reference. Assignments were then updated using Kozloff (1987), a general West coast reference. Most taxa collected are found on both the East and West coasts, and family-level taxonomy has not changed much in the last few decades.

Richness ( $S$ ) was the number of taxa (families) per station. Diversity was Simpson’s  $D$  calculated using:

$$D = 1/\sum p_i^2$$

where  $p_i$  is the abundance of the  $i$ th taxon as a proportion of total abundance.  $D$  is the number of “dominant taxa”, with higher values indicating greater diversity. Simpson’s evenness ( $E$ ) is then  $D/S$ , or the number of dominant taxa relative to the total number of taxa. Although evenness is calculated from diversity, diversity is defined as a function of richness and evenness (i.e.,  $D = S \times E$ ).

Non-metric multidimensional scaling (NMDS) was used to derive summary community composition measures. NMDS can be considered a non-parametric analogue of PCA; Clarke (1993) discusses methods and applications. First abundances of each taxon (family) were expressed as a percent of total abundance. Second, Bray-Curtis (B-C) similarities were calculated between all possible pairs of stations. These B-C similarities are the percentage of invertebrates shared between stations (percent similarity). Third, B-C similarities were subjected to NMDS. NMDS iteratively finds the  $k$ -dimensional solution (i.e., set of axes) that best reproduces the original pair-wise similarity matrix. The stress coefficient, which ranges from 0 (perfect fit to original matrix) to 1 (no fit), can be used to assess the adequacy of the NMDS solution. Positions of stations along the

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<sup>5</sup> In some environments, usually neashore, nemertean and oligochaetes can make some contribution to standing crop when they are abundant and larger organisms (for instance, echinoderms) are rare or absent.

NMDS axes (Multidimensional Scores, MDS1, MDS2 etc.) can then be used as summary measures for further analyses. In SYSTAT (the statistical software used for NMDS), NMDS solutions and axes are rotated so that variance is greatest along MDS1 (i.e., MDS1 is the major axis of variance). All stations sampled in 2000, 2004 and 2005 were included in the NMDS, since all stations were included in some analysis of MDS scores.

### **Statistical Analysis**

Summary statistics for invertebrate community variables were calculated over all 44 stations sampled in 2005. Rank correlations ( $r_s$ ) among the variables were also calculated. Relative abundances of major taxa, and absolute abundances (numbers) of Echinodermata (a rarer taxon of potential interest) were included in these two steps, but not in analyses described below.

Rank correlations between invertebrate community variables and sediment physical and chemical characteristics were also calculated for 2005 samples, although in-depth analyses of these relationships was conducted as part of the Integrated Assessment of data from all three sample years (Section 5.3.5). Benthic community variable values for the two stations with low amphipod survival in toxicity tests were also tabulated, to determine if the laboratory effects were associated with field effects.

Rank-rank distance-depth relationships were analyzed, followed by more specific parametric regression analysis when warranted. The RM regression model described in Section 5.3.1.2 was used to compare invertebrate community variables among years. Threshold distances (in this case referred to as zones of effects rather than zones of influence) for selected variables were compared between 2004 and 2005.

Total and amphipod abundances were log-transformed for parametric analyses. A  $\log_{10}(Y+1)$  transformation was used for amphipod abundance because some samples contained no (0) amphipods.

### **5.3.5 Integrated Assessment**

A dose-response approach, analyzing relationships between invertebrate community variables (biological response or Y) and sediment physical and chemical characteristics (dose or X), was used for the integrated assessment of the two major SQT components over the three sample years (2000, 2004, 2005). The dose-response approach assessed the effects of X variables other than distance, used all stations sampled in each year rather than the subset of stations sampled in all years, and often relied on non-parametric rather than parametric analyses. The relative merits of the dose-response versus RM/distance approach are further discussed below and in Appendix B-5. The two approaches should be regarded as complementary, addressing a variety of questions that cannot be addressed via a single analysis.

Toxicity was not included in the analyses, because only one (of 146) samples was toxic to amphipods in three sample years and no samples were toxic to luminescent bacteria. Depth was included in some analyses. Depth measurements in 2000 were referenced to a different "0" depth point than 2004 measurements. Therefore, 2000 depths for the nine stations not sampled in 2004 were converted to 2004 depth equivalents based on a regression relationship between 2000 and 2004 depths for the 37 stations sampled in

both years. All stations sampled in 2005 were sampled in 2004, so 2004 depths were used for stations sampled in 2005.

The dose-response relationships analyzed were:

- relationships between community variables and sediment particle size (% fines and gravel) and TOC (primarily an assessment of natural habitat effects and variance); and
- relationships between community variables and drilling mud tracers (barium, >C<sub>10</sub>-C<sub>21</sub> HCs) (project-related dose-response relationships).

Particle size and TOC effects on invertebrate communities were assessed because they are important sources of natural variance in other marine monitoring programs, including the nearby Terra Nova EEM program (Petro-Canada 2005). Depth was included in these analyses to address the possibility that particle size and depth effects may be correlated or confounded.

The first step in analyses of natural (particle size, TOC and depth) effects was to calculate rank correlations ( $r_s$ ) between community  $Y$  variables and  $X$  variables of interest within each year. The bivariate correlations were then compared among years (blocks) using the van Belle tests (also known as the Mann-Kendall or M-K test) described in Appendix B-5. The van Belle test first tests for differences in correlations among years, and then tests the common or average correlation over all years if there are no differences among years. Rank-rank regressions of selected invertebrate community variables on both particle size and depth (i.e., partial correlation/regression analyses) were then compared in Analysis of Covariance (ANCOVA).

Using tracers, rather than distance variables, as  $X$  variables in dose-response relationships addressed some problems with analysis of distance effects. The spatial distribution of tracer concentrations will incorporate directional and other non-distance and localized project effects, especially around individual drill centres. A single tracer  $X$  variable may be a simpler and better predictor of community  $Y$  variable values than one or more distance  $X$  variables. Threshold tracer concentrations below which effects do not occur may also be of interest.

As was done for analysis of effects of particle size, TOC and depth, the first step was to compare rank correlations for invertebrate community variables ( $Y$ ) versus barium and >C<sub>10</sub>-C<sub>21</sub> HCs ( $X$ ) among the three sample years using van Belle tests. These tests should be regarded as coarse screening because ranges of tracer values, especially for >C<sub>10</sub>-C<sub>21</sub> HCs, were much wider in 2004 and 2005 than in 2000. Parametric bivariate and hockey-stick relationships between selected community variables ( $Y$ ) and >C<sub>10</sub>-C<sub>21</sub> HC concentrations ( $X$ ) for 2004 and 2005 were also calculated and compared between the two years. Barium concentrations were not a useful  $X$  variable for the parametric regressions because most barium concentrations in 2004 and 2005 were within the narrow baseline (2000) range of 120 to 210 mg/kg, where  $Y$  variables varied widely and effects attributable to elevated barium concentrations from drilling could not easily be determined.

## 5.4 Results

### 5.4.1 Physical and Chemical Characteristics

Table 5-4 provides summary statistics for sediment physical and chemical characteristics occurring at or above EQL at one or more stations in 2000, 2004 and 2005. All variables measured on sediment are provided in Table 5-3. Toluene was detected at levels close to EQL (0.03) in one sample in 2005 and was not detected in previous years.  $>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HCs have been detected in 2004 and 2005, but not in 2000. With the exception of Naphthalene, which was detected in 2000, PAHs have never been detected in sediment samples. Commonly detected metals in all three sampling years include: aluminum, barium, chromium, iron, lead, manganese, strontium, uranium, vanadium and zinc.

**Table 5-4 Summary Statistics for Physical and Chemical Characteristics (2000, 2004, 2005)**

Variable	Year	n	n<EQL	Min	Max	Median	Mean	SD	CV
Toluene	2000	46	46	<0.025	<0.025	<0.025			
	2004	56	56	<0.025	<0.025	<0.025			
	2005	44	43	<0.03	0.040	<0.03			
$>C_{10}-C_{21}$	2000	46	46	<0.25	<0.25	<0.25			
	2004	56	11	<0.25	275.000	0.740			
	2005	44	5	0.3	260.00	1.00			
$>C_{21}-C_{32}$	2000	46	46	<0.25	<0.25	<0.25			
	2004	56	45	<0.25	0.920	<0.25			
	2005	44	19	0.30	1.70	0.30			
Naphthalene	2000	46	45	<0.05	0.070	<0.05			
	2004	56	56	<0.05	<0.05	<0.05			
	2005	44	44	<0.05	<0.05	<0.05			
Total Carbon	2000	46	0	0.70	1.30	1.00	0.99	0.12	12
	2004	56	0	0.70	1.40	1.05	1.05	0.12	11
	2005	44	0	0.90	1.70	1.00	1.07	0.15	14
Total Inorganic Carbon	2000	46	6	<0.1	0.40	0.10			
	2004	56	52	<0.3	0.50	<0.3			
	2005	44	24	<0.2	0.72	<0.2			
Total Organic Carbon	2000	46	0	0.60	1.00	0.90	0.85	0.09	11
	2004	56	0	0.60	1.20	0.95	0.94	0.10	11
	2005	44	0	0.60	1.10	0.90	0.89	0.09	10
Aluminum	2000	46	0	6400	11000	8250	8243	651	8
	2004	56	0	6500	9500	8300	8173	709	9
	2005	44	0	5700	14000	8350	8502	1122	13
Arsenic	2000	46	33	<2	2.0	<2			
	2004	56	56	<2	<2	<2			
	2005	44	44	<2	<2	<2			
Barium	2000	46	0	120.0	210.0	160.0	163.7	19.4	12
	2004	56	0	110.0	1400.0	160.0	203.4	177.7	87
	2005	44	0	93.0	810.0	170.0	210.5	116.2	55

Variable	Year	n	n<EQL	Min	Max	Median	Mean	SD	CV
Cadmium	2000	46	46	<0.05	<0.05	<0.05			
	2004	56	38	<0.05	0.080	<0.05			
	2005	44	35	<0.05	0.070	<0.05			
Chromium	2000	46	0	3.0	4.0	3.0	3.5	0.5	15
	2004	56	0	3.0	7.0	4.0	3.8	0.7	18
	2005	44	0	2.8	5.5	3.55	3.7	0.6	16
Cobalt	2000	46	44	<1	1.0	<1			
	2004	56	50	<1	1.0	<1			
	2005	44	44	<1	<1	<1			
Copper	2000	46	41	<2	4.0	<2			
	2004	56	19	<2	3.0	<2			
	2005	44	40	<2	2.9	<2			
Iron	2000	46	0	1100	2300	1400	1461	244	17
	2004	56	0	850	2400	1500	1489	315	21
	2005	44	0	1100	2900	1600	1677	399	24
Lead	2000	46	0	2.10	5.10	2.70	2.79	0.44	16
	2004	56	0	2.00	4.00	2.75	2.75	0.33	12
	2005	44	0	1.8	5.9	2.8	2.98	0.63	21
Lithium	2000	46	46	<5	<5	<5			
	2004	56	31	<2	2.0	<2			
	2005	44	44	<2	<2	<2			
Manganese	2000	46	0	25.0	70.0	36.0	38.7	10.1	26
	2004	56	0	17.0	82.0	38.0	40.1	12.7	32
	2005	44	0	22.0	96.0	40.5	45.6	16.1	35
Nickel	2000	46	44	<2	2.0	<2			
	2004	56	54	<2	2.0	<2			
	2005	44	43	<2	2	<2			
Strontium	2000	46	0	37.0	60.0	47.0	47.5	3.5	7
	2004	56	0	34.0	64.0	46.0	47.0	4.9	10
	2005	44	0	30.0	75.0	49.0	49.2	6.4	13
Thallium	2000	46	1	<0.1	0.10	0.10			
	2004	56	0	0.10	0.10	0.10	0.10	0.00	0
	2005	44	40	<0.1	0.12	<0.1			
Uranium	2000	46	0	0.20	0.30	0.20	0.20	0.02	10
	2004	56	0	0.20	0.30	0.20	0.21	0.02	11
	2005	44	0	0.13	0.29	0.21	0.22	0.04	17
Vanadium	2000	46	0	5.0	8.0	6.0	6.4	0.7	11
	2004	56	0	4.0	7.0	6.0	5.7	0.8	13
	2005	44	0	4.5	9.2	5.7	5.8	0.9	16
Zinc	2000	46	0	4.0	14.0	6.0	6.4	2.3	35
	2004	56	10	<5	9.0	<5			
	2005	44	0	4.9	10.0	7.1	7.0	1.1	15
Ammonia	2000	NA							
	2004	56	0	2.17	64.60	7.10	9.23	9.00	98
	2005	44	0	2.30	49.0	7.25	8.49	7.16	84

Variable	Year	n	n<EQL	Min	Max	Median	Mean	SD	CV
Sulphide	2000	NA							
	2004	56	53	<2	3.0	<2			
	2005	44	31	<0.2	1.00	<0.2			
Sulphur	2000	NA							
	2004	56	1	<0.02	0.080	0.030			
	2005	44	1	<0.02	0.048	0.025			
Moisture	2000	46	0	14.00	22.00	19.00	18.46	1.56	8
	2004	56	0	16.00	23.00	18.00	18.50	1.49	8
	2005	44	0	17.00	20.00	18.00	18.36	0.89	5
% Clay	2000	46	0	0.29	0.83	0.62	0.61	0.12	20
	2004	56	0	0.14	1.02	0.61	0.60	0.17	28
	2005	44	0	0.01	1.14	0.565	0.58	0.22	38
% Gravel	2000	46	0	0.00	2.30	0.55	0.67	0.54	81
	2004	56	0	0.00	5.60	0.80	1.09	1.09	100
	2005	44	0	0.00	11.2	0.65	1.32	1.94	146
% Sand	2000	46	0	96.63	99.12	98.46	98.32	0.55	1
	2004	56	0	92.62	98.59	97.64	97.35	1.21	1
	2005	44	0	87.74	98.98	98.09	97.45	1.91	2
% Silt	2000	46	0	0.15	0.94	0.39	0.42	0.14	34
	2004	56	0	0.47	2.41	0.88	0.95	0.37	39
	2005	44	0	0.12	1.81	0.635	0.65	0.31	48

Note: - All units are mg/kg except where indicated  
 - 2000 data exclude the two remote Reference stations; Ammonia, sulphur and sulphides were not measured in 2000  
 - Statistics are reported to one more significant digit than what is given for EQL (see Table 5-3)

5.4.1.1 Correlations Within and Among Groups of Variables (2005)

Sediments sampled in 2005 (and previous years) were predominantly (usually more than 90%) sand (Table 5-4). One or both of the “non-sand” components, gravel and fines, was expected to be negatively correlated with sand content, since percentages of the three particle size categories sum to 100%. Gravel content, which was usually the major non-sand component by weight and varied among stations from 0 to 11.2%, was strongly negatively correlated with sand content (Table 5-5). Fines content varied over a narrow range (0.77 to 2.67%), and was uncorrelated with gravel and sand content. Based on these correlations, sand and gravel content were considered redundant, and sand content eliminated from further analyses.

Table 5-5 Spearman Rank Correlations (*r<sub>s</sub>*) Among Particle Size Categories and TOC (2005)

	% fines	% sand	% gravel
% sand	-0.157		
% gravel	-0.069	<b>-0.948***</b>	
TOC	<b>0.520***</b>	0.165	-0.292

Note: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)

TOC levels in sediments collected in 2005 were low (0.6 to 1.1 g/kg) and did not vary widely among stations. Despite the limited variance in TOC, values were significantly

positively correlated with fines content (Table 5-5). Organic matter (i.e., TOC) should be associated with finer particles, although the expected positive correlation between the two variables has not always been significant (e.g., as in 2004; Husky Energy 2005) over the narrow range of TOC and fines values in the White Rose area.

Concentrations of the two primary drilling mud tracers, barium and  $>C_{10}-C_{21}$  HCs, were strongly and significantly positively correlated (Table 5-6). This correlation was expected since both WBMs and SBMs were used at all three drill centres and barium is a constituent of both types of muds.  $>C_{21}-C_{32}$  HC concentrations were positively correlated with concentrations of both barium and  $>C_{10}-C_{21}$  HCs, indicating that detectable  $>C_{21}-C_{32}$  HC concentrations were more likely to occur where concentrations of the two primary tracers were high.  $>C_{21}-C_{32}$  HCs were not included in further analyses, because of the strong correlation with  $>C_{10}-C_{21}$  HCs and the fact that the latter were more suitable for most analyses (i.e., with more values greater than EQL).

**Table 5-6 Spearman Rank Correlations ( $r_s$ ) Among Barium and HC Concentrations (2005)**

	Barium	$>C_{10}-C_{21}$ HCs
$>C_{10}-C_{21}$ HCs	<b>0.823***</b>	
$>C_{21}-C_{32}$ HCs	<b>0.519***</b>	<b>0.668***</b>

Note: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)

Concentrations of the nine other frequently detected metals in sediments collected in 2000, 2004 and 2005 were positively correlated with each other and with the first Principal Component (Metals PC1) derived from these concentrations (Table 5-7). Metals PC1 accounted for more than 50% of the total variance among the 146 samples and was used as a summary measure of “total” metals concentrations for further analyses. The secondary axes of variance (PC2 and PC3) accounted for minimal variance and were not further analyzed.

**Table 5-7 Correlations ( $r$ ) Between Concentrations of Frequently Detected Metals and PCs Derived from those Concentrations (2000, 2004, 2005)**

Metal	Correlation ( $r$ ) with:		
	PC1	PC2	PC3
Iron	<b>0.902</b>	0.281	-0.074
Aluminum	<b>0.883</b>	-0.200	0.068
Strontium	<b>0.876</b>	-0.306	0.147
Manganese	<b>0.853</b>	0.368	-0.085
Vanadium	<b>0.705</b>	0.256	-0.219
Chromium	<b>0.699</b>	0.329	0.009
Lead	<b>0.646</b>	<b>-0.622</b>	0.113
Uranium	<b>0.616</b>	0.088	<b>0.587</b>
Zinc	<b>0.586</b>	-0.318	<b>-0.573</b>
% variance	57.9	11.3	8.6

Notes: - Metals are listed in descending order of their correlation with PC1  
 -  $|r| \geq 0.5$  in bold  
 - Concentrations were  $\log_{10}$  transformed prior to deriving PC  
 -  $n = 146$  stations; 44 in 2005, 56 in 2004, 46 in 2000

Metals PC1 scores were weakly but significantly positively correlated with barium concentrations, which will naturally co-vary with concentrations of other metals (e.g., as

in 2000; Husky Oil 2001) (Table 5-8). However, the natural covariance between barium concentrations and concentrations of other metals was small relative to project-related covariance of barium and >C<sub>10</sub>-C<sub>21</sub> HC concentrations (compare correlations in Table 5-8). Metals PC1 scores were uncorrelated with the other drilling mud tracer, >C<sub>10</sub>-C<sub>21</sub> HCs.

**Table 5-8 Spearman Rank Correlations (*r<sub>s</sub>*) Among Chemistry Variables (2005)**

	Barium	>C <sub>10</sub> -C <sub>21</sub> HCs	Metals PC1	Ammonia	Sulphur
>C <sub>10</sub> -C <sub>21</sub> HCs	<b>0.823***</b>				
Metals PC1	0.335*	0.105			
Ammonia	0.032	-0.159	0.127		
Sulphur	0.339*	0.333*	0.110	-0.180	
Redox	-0.501**	<b>-0.565***</b>	-0.013	0.208	-0.479**

Note: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)

Ammonia levels were uncorrelated with tracer, metal, sulphur and redox levels (Table 5-8). Sulphur and redox levels were negatively correlated with each other. Sulphur levels increased, and redox levels decreased, with increasing tracer concentrations, suggesting that drilling and drill cutting discharges may have elevated sulphur (presumably sulphate) concentrations and reduced redox levels.

Metals PC1 scores and ammonia concentrations were weakly but significantly positively correlated with sediment fines and TOC content (Table 5-9). Tracer, sulphur and redox levels were uncorrelated with fines and TOC. Stronger correlations between metals, HCs and finer organic particles would normally be expected, but fines and TOC levels were low and varied little among stations.

**Table 5-9 Spearman Rank Correlations (*r<sub>s</sub>*) Between Chemistry Variables, Fines and TOC (2005)**

Chemistry variable	Correlation ( <i>r<sub>s</sub></i> ) with:	
	% fines	TOC
Barium	0.281	0.130
>C <sub>10</sub> -C <sub>21</sub> HCs	0.088	-0.034
Metals PC1	0.356*	0.302*
Ammonia	0.361*	0.304*
Sulphur	0.173	0.067
Redox	-0.298	0.054

Note: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)

**5.4.1.2 Depth and Distance Effects (2005)**

Table 5-10 provides results of rank-rank regressions of sediment physical and chemical characteristics on depth and distances from the drill centres. Overall multiple correlations (*R*) for regression models with more than one *X* variable can range from 0 to 1. Partial correlations (*r*) for each *X* variable can range from -1 to 1, and provide the correlation between each *X* variable and *Y* with the effects of other *X* variables held constant or removed.



**Table 5-10 Results of Rank-Rank Regressions of Physical and Chemical Characteristics on Depth and Distances from the Drill Centres (2005)**

Y Variable	X=Depth & distances from each drill centre					X=Depth & distance from nearest drill centre (Min <i>d</i> )			X=Depth	X=Min <i>d</i>
	Overall <i>R</i>	Partial <i>r</i>				Overall <i>R</i>	Partial <i>r</i>		<i>r<sub>s</sub></i>	<i>r<sub>s</sub></i>
		Depth	N <i>d</i>	C <i>d</i>	S <i>d</i>		Depth	Min <i>d</i>		
Barium	<b>0.704***</b>	0.058	-0.149	-0.179	-0.306	<b>0.775***</b>	0.231	<b>-0.774***</b>	-0.071	-
>C <sub>10</sub> -C <sub>21</sub> HCs	<b>0.891***</b>	-0.435**	-0.286	-0.214	<b>-0.572***</b>	<b>0.905***</b>	-0.247	<b>-0.890***</b>	-0.358*	<b>0.760***</b>
% fines	0.485*	0.312*	0.129	0.072	-0.203	0.387*	0.386*	-0.146	0.362*	-0.028
% gravel	0.318	0.301	0.029	-0.184	0.182	0.301	0.283	-0.187	0.240	-0.106
TOC	0.196	0.024	-0.167	0.141	-0.127	0.093	0.084	0.014	0.092	0.039
Metals PC1	0.293	0.249	-0.005	-0.119	0.020	0.211	0.211	-0.055	0.204	0.006
Ammonia	<b>0.746***</b>	<b>0.523***</b>	-0.253	0.451**	<b>-0.506***</b>	<b>0.639***</b>	<b>0.633***</b>	-0.097	<b>0.634***</b>	0.108
Sulphur	0.408	-0.131	0.234	-0.142	-0.029	0.287	-0.144	-0.205	-0.205	-0.251
Redox	<b>0.675***</b>	0.086	-0.376*	0.222	0.199	0.443*	0.051	0.416**	0.169	0.441**

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - N, C and S *d* = distances from the Northern, Central and Southern drill centres  
 - Min *d* = distance from the nearest drill centre  
 - All Y and X variables were rank-transformed

### Tracers

Barium and >C<sub>10</sub>-C<sub>21</sub> HC concentrations decreased significantly with distances from the drill centres (negative *r* or *r<sub>s</sub>* in Table 5-10). For both tracers, multiple *R* were higher when distance from the nearest drill centre was used as a single distance X variable than when distances from each drill centre were used as separate X variables, suggesting that effects from all drill centres were equal. As discussed below and elsewhere, differences in contamination among drill centres were localized, usually evident only in the immediate vicinity of the drill centres.

Barium concentrations were uncorrelated with depth, but >C<sub>10</sub>-C<sub>21</sub> HC concentrations decreased significantly with increasing depth (Table 5-10). In the long term, finer particles, including cuttings, would be expected to accumulate at greater depths due to down-slope movement. There was little or no evidence from the multi-year analyses (Section 5.4.1.3) for depth effects on >C<sub>10</sub>-C<sub>21</sub> HCs, and depth effects on >C<sub>10</sub>-C<sub>21</sub> HCs were not considered in analyses of distance effects provided below.

In parametric models for barium and >C<sub>10</sub>-C<sub>21</sub> HCs, using distances from each drill centre as opposed to distance from the nearest drill centre increased error variances (i.e., treating the drill centres separately added “noise”). However, adding a threshold distance value (*X<sub>T</sub>*) to regressions of both variables on distance from the nearest drill centre significantly reduced error variances relative to bivariate regressions (Table 5-11). The hockey-stick regressions also reduced (but did not eliminate) the effects of extreme values and some violations of assumptions of parametric analyses.

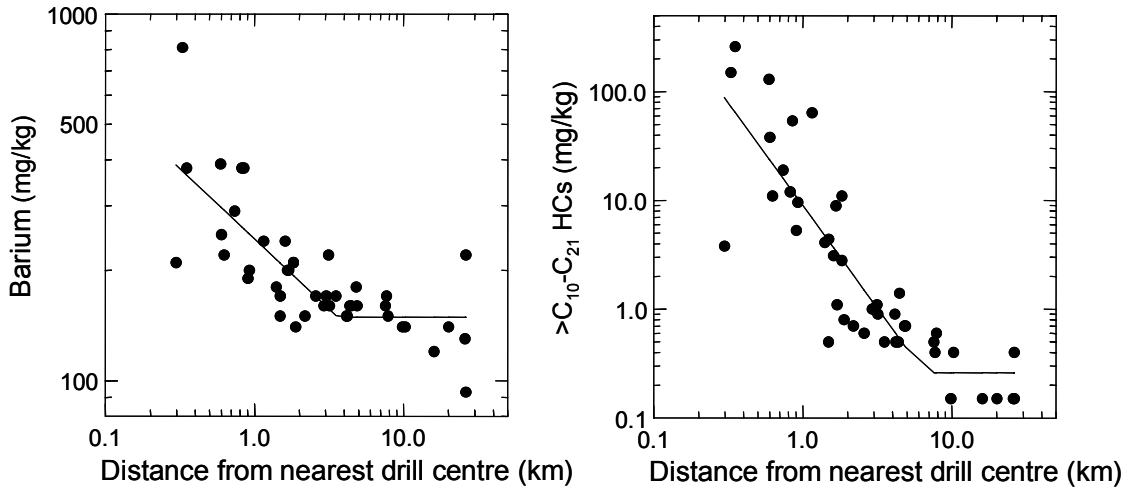
**Table 5-11 Results for Parametric Distance Models for Barium, >C<sub>10</sub>-C<sub>21</sub> HCs and Redox (2005)**

Result/Estimate	Barium (mg/kg)	>C <sub>10</sub> -C <sub>21</sub> HCs (mg/kg)	Redox (mV)
Regression on distance from nearest drill centre			
<i>r</i>	<b>-0.723***</b>	<b>-0.855***</b>	0.464**
Full distance model			
Overall <i>R</i>	<b>0.714***</b>	<b>0.837***</b>	<b>0.701***</b>
<i>p</i> for all drill centres vs nearest	>1 <sup>1</sup>	>1 <sup>1</sup>	<b>&lt;0.001</b>
Hockey-stick model			
Overall <i>R</i>	<b>0.767***</b>	<b>0.875***</b>	<b>0.547***</b>
<i>p</i> for adding threshold ( <i>X<sub>T</sub></i> )	0.015	0.016	0.033
antilog <i>a</i> (blade or background <i>Y</i> value)	149	0.26	256
95% CI	131 to 169	0.14 to 0.49	241 to 271
<i>b</i> (slope of shaft)	-0.383	-1.89	0.135
95% CI	-0.520 to -0.246	-2.32 to -1.46	0.036 to 0.235
antilog <i>X<sub>T</sub></i> (threshold distance in km)	3.6	6.5	2.6
95% CI	2.1 to 6.3	3.9 to 10.8	1.0 to 6.4

- Notes:
- \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)
  - *X* variables for the full distance model were distances from each drill centre
  - The *X* variable for the hockey-stick model was distance from the nearest drill centre
  - Both models were compared to bivariate regressions of *Y* on distance to the nearest drill centre
  - All *Y* and *X* variables were log-transformed
  - <sup>1</sup>—Using distances from each drill centre as opposed to the nearest drill centre as *X* increased error variances

Table 5-11 provides parameter estimates for the hockey-stick relationships, which are the lines used in Figure 5-6<sup>6</sup>. Estimates of threshold distances were zones of influence. Barium concentrations reached estimated background levels (149 mg/kg) at 3.6 km from the nearest drill centre. >C<sub>10</sub>-C<sub>21</sub> HC concentrations reached background levels (effectively EQL of 0.3 mg/kg) at 6.5 km. Thus, >C<sub>10</sub>-C<sub>21</sub> HC contamination was spatially more extensive than barium contamination. The distance gradient (slope of the shaft) was also steeper for >C<sub>10</sub>-C<sub>21</sub> HCs. For both tracers, variance at the lowest distances was greater than at intermediate distances. These are the stations at which differences among drill centres (“which drill centre is nearest?”) are important. Variance (presumably natural) about the hockey-stick model for barium also increased for more remote stations beyond the estimated distance threshold, a common occurrence for many variables. Most parametric distance models fit intermediate distances best.

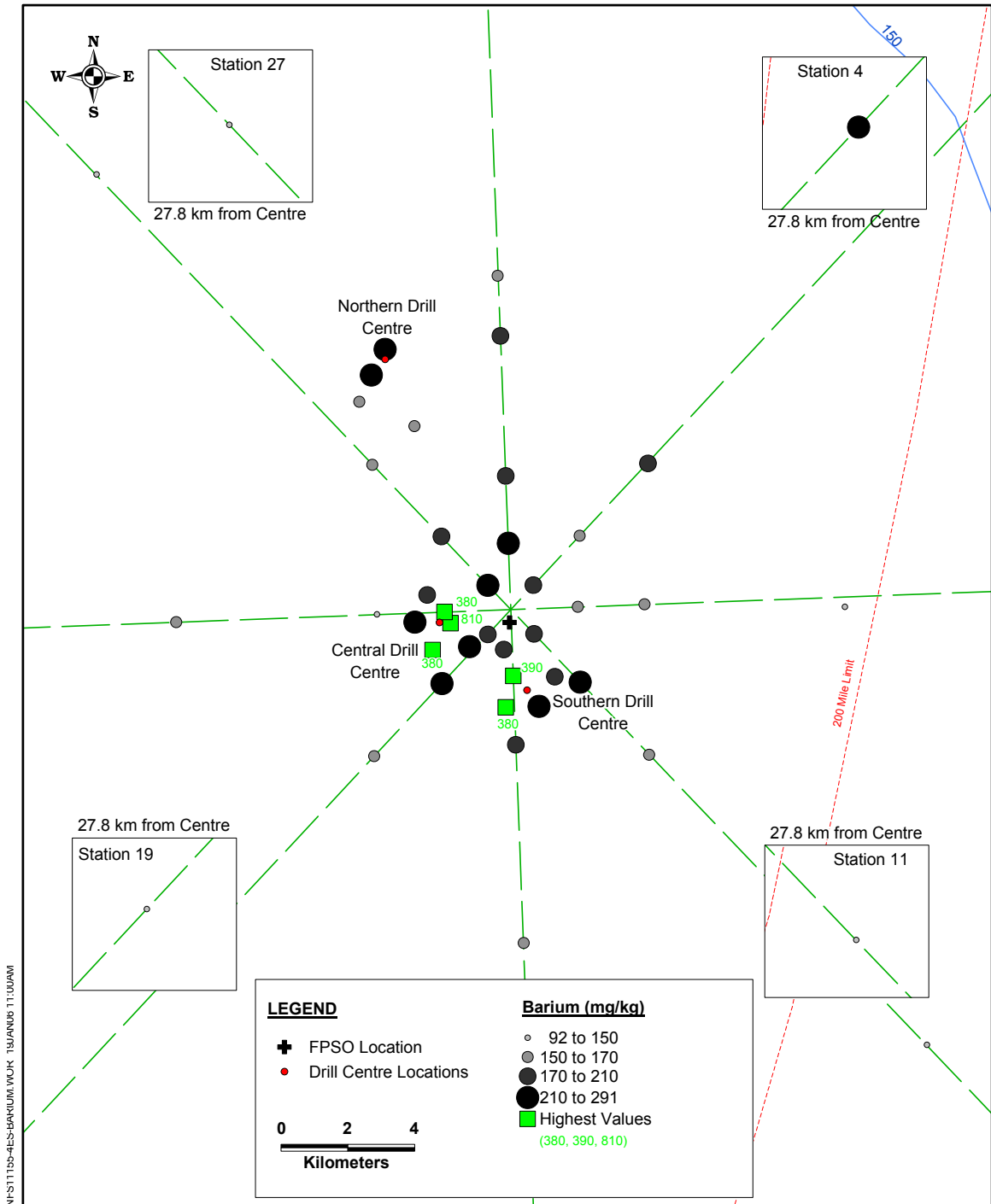
<sup>6</sup> Here and elsewhere in Section 5.4, relationships between physical, chemical and biological *Y* variables and depth, distance and tracer concentrations (*X* variables) were not plotted, unless the relationships, or changes in relationships over time, were significant at *p* ≤ 0.05.



**Figure 5-6 Barium and >C<sub>10</sub>-C<sub>21</sub> HCs versus Distance from the Nearest Drill Centre (2005)**

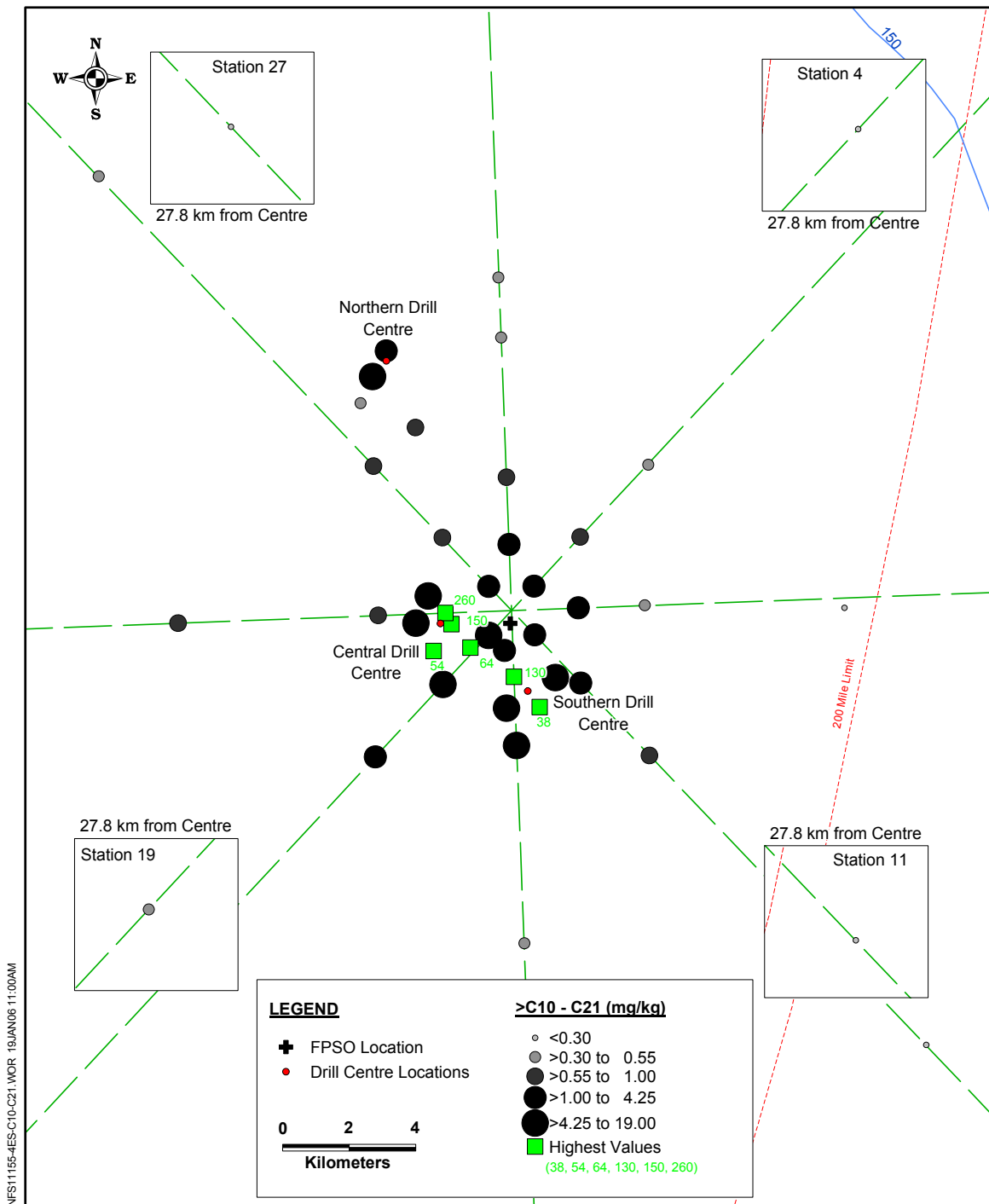
Figure 5-7 plots the spatial distribution of barium. Concentrations were greatest at several stations within 1 km of the Central and Southern drill centres, and also elevated above background at the two stations nearest the Northern drill centre. Concentrations were greater to the south and east around the Central and Southern drill centres than to the north or west.

The spatial distribution of >C<sub>10</sub>-C<sub>21</sub> HCs was similar but with more extensive contamination (effectively any concentration above EQL of 0.3 mg/kg) (Figure 5-8). Concentrations around the Central and Southern drill centres were greater to the southeast than in other directions. >C<sub>10</sub>-C<sub>21</sub> HC concentrations around the two drill centres were similar, whereas barium concentrations were greater around the Central drill centre (compare Figures 5-7 and 5-8). There did not appear to be any decrease in >C<sub>10</sub>-C<sub>21</sub> HC concentrations from southwest to northeast along the gradient of increasing depth; distance effects overwhelmed any depth effects.



**Figure 5-7 Spatial Distribution of Barium (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software

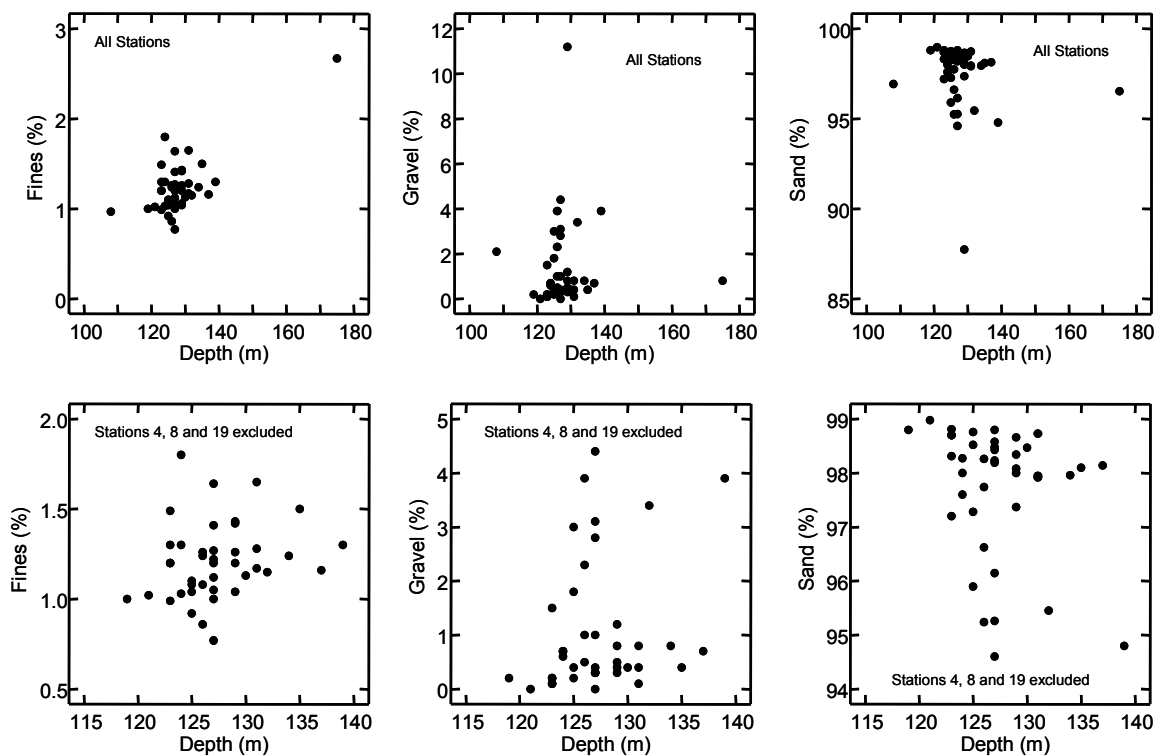


**Figure 5-8 Spatial Distribution of >C<sub>10</sub>-C<sub>21</sub> HCs (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software

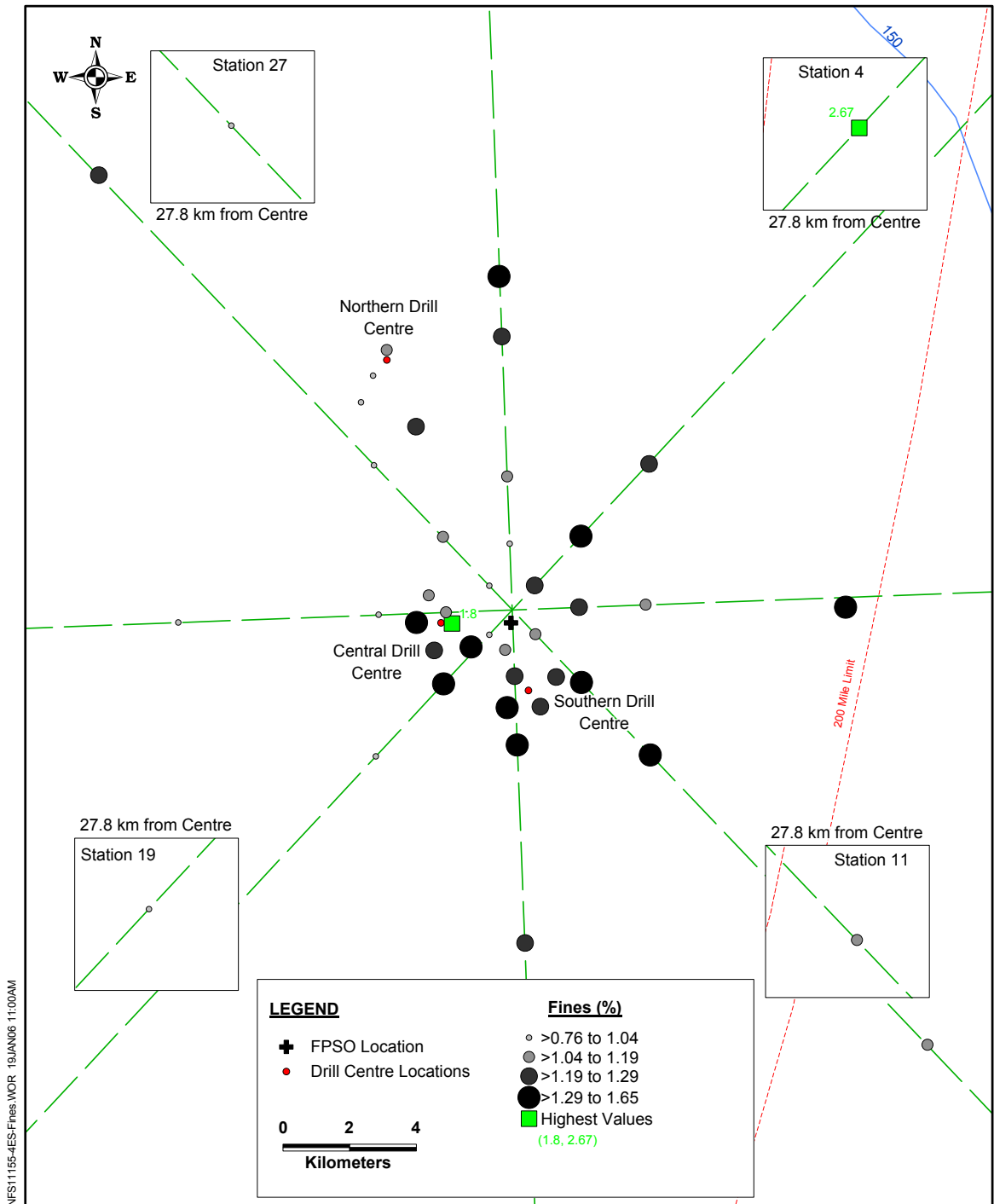
**Particle Size and TOC**

Over all stations, fines and, to a lesser extent, gravel content increased with increasing depth (Table 5-10; Figure 5-9). Fines and gravel content were uncorrelated with distances from the drill centres. Figure 5-10 provides the spatial distribution of fines content. The highest value (2.67% versus less than 2% at other stations) occurred at station 4, the Northeast reference station and the deepest station (depth = 175 m). Fines content was also low at station 19, the Southwest reference station and the shallowest station (depth = 108 m). The highest gravel content (11.2% versus less than 5% at other stations) occurred at station 8, an intermediate station in terms of depth and distances from drill centres. Fines content at station 8 was low (approximately 1%). With these three extreme stations eliminated, correlations between fines and gravel content versus depth were not significant (although they were still positive) (Figure 5-9; bottom row); and correlations with distances remained weak and not significant.



**Figure 5-9 Fines, Gravel and Sand Content versus Depth (2005)**

TOC was uncorrelated with distances and depth (Table 5-10), probably because spatial variance of TOC was minimal.

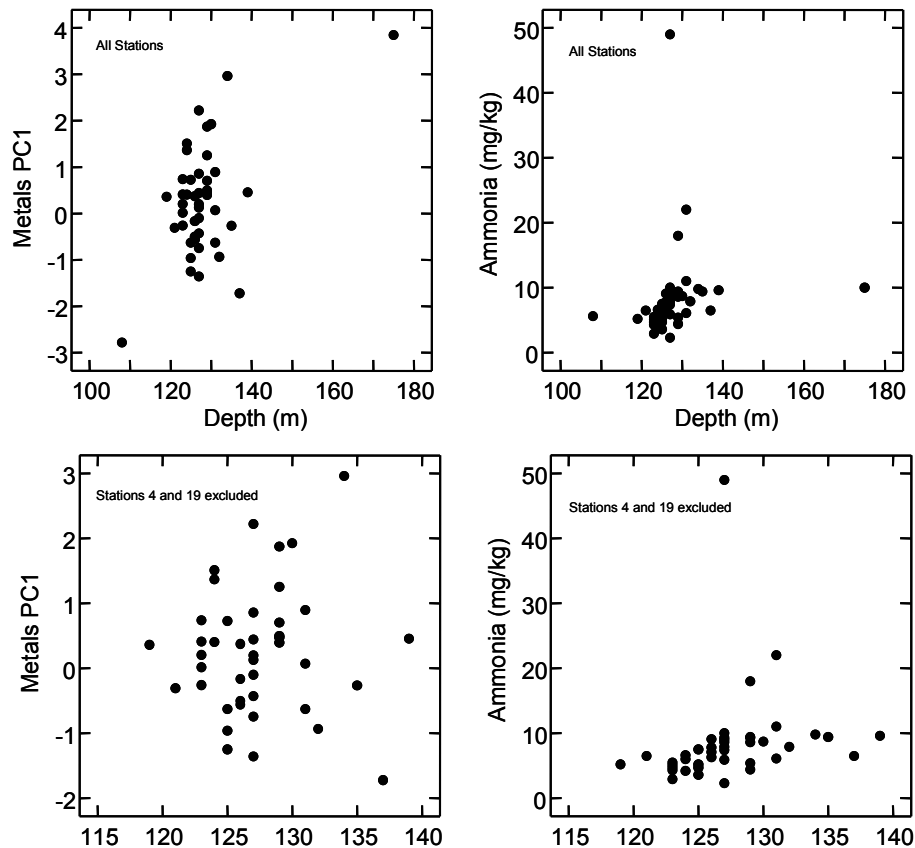


**Figure 5-10 Spatial Distribution of % Fines (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software

**Metals**

Concentrations of metals other than barium (i.e., Metals PC1 scores) were positively, but not significantly, correlated with depth, and uncorrelated with distances from the drill centres. The depth correlation was largely a function of the extreme Metals PC1 and depth values for reference stations 4 and 19, and much weaker with these two stations excluded (Figure 5-11). More generally, correlations between Metals PC1 and depth for any set or subset of stations were weaker versions of depth correlations for fines content, since metal and fines levels were positively correlated (Table 5-9).

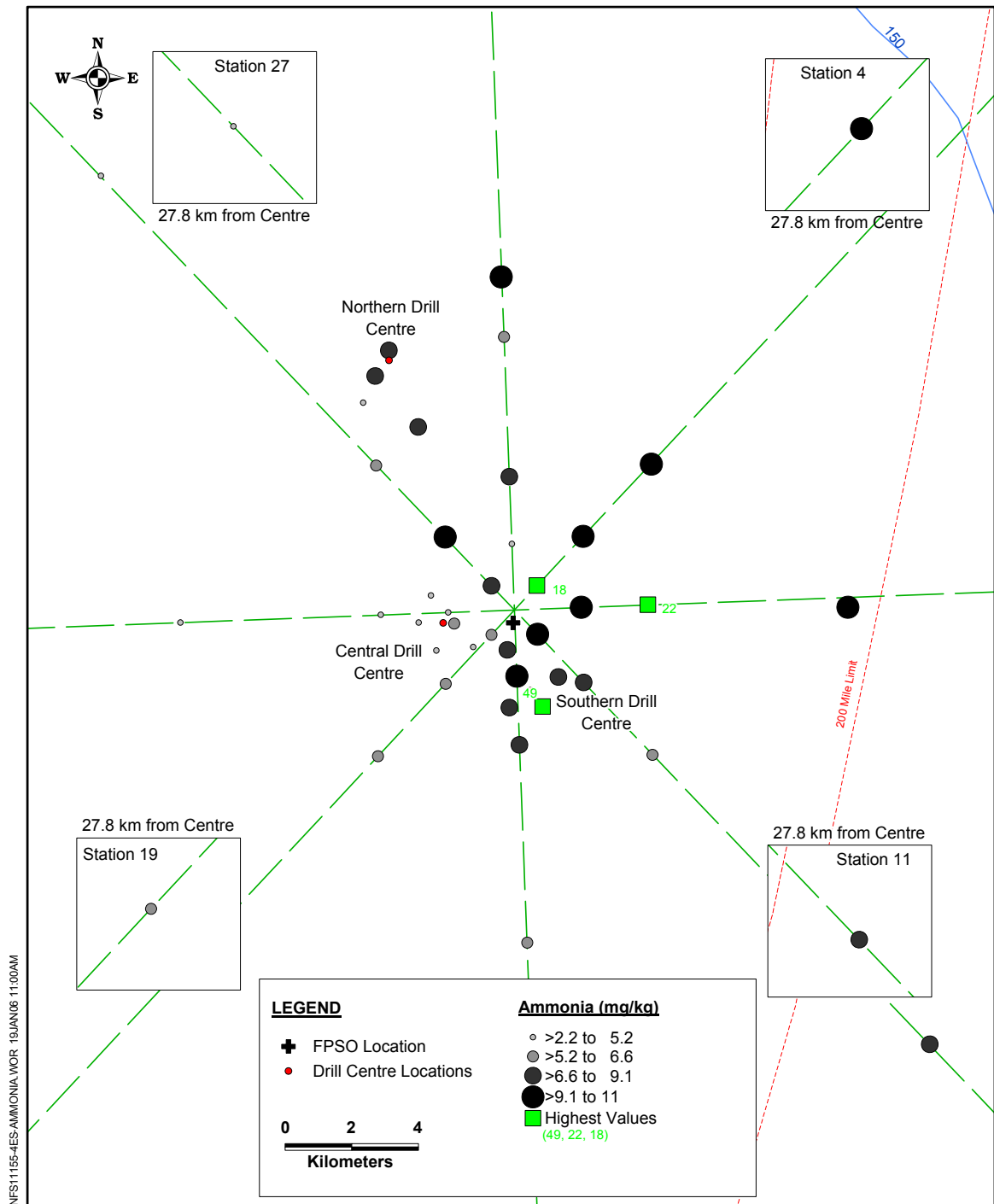


**Figure 5-11 Metals PC1 and Ammonia versus Depth (2005)**

**Ammonia, Sulphur and Redox**

Ammonia levels were significantly positively correlated (i.e., increased) with depth (Table 5-10; Figure 5-11), and generally increased from southwest to northeast (the direction of the depth gradient) (Figure 5-12). The ammonia concentration at station S1 was anomalously high (49 mg/kg versus less 25 mg/kg at other stations). Station S1 was near (0.6 km from) the Southern drill centre, but there was no evidence that ammonia levels were elevated at other stations within 1 km of drill centres (Figure 5-12), and no overall relationship between ammonia and distances (Table 5-10). Even with stations 4 and 19 (extreme depths) excluded, and station S1 (extreme ammonia value) included, the rank correlation between ammonia and depth remained strongly and significantly positive ( $r_s = 0.614$ ;  $p \leq 0.001$ ) (Figure 5-11, bottom plot; ranking substantially reduced the effects of the extreme ammonia value at S1).

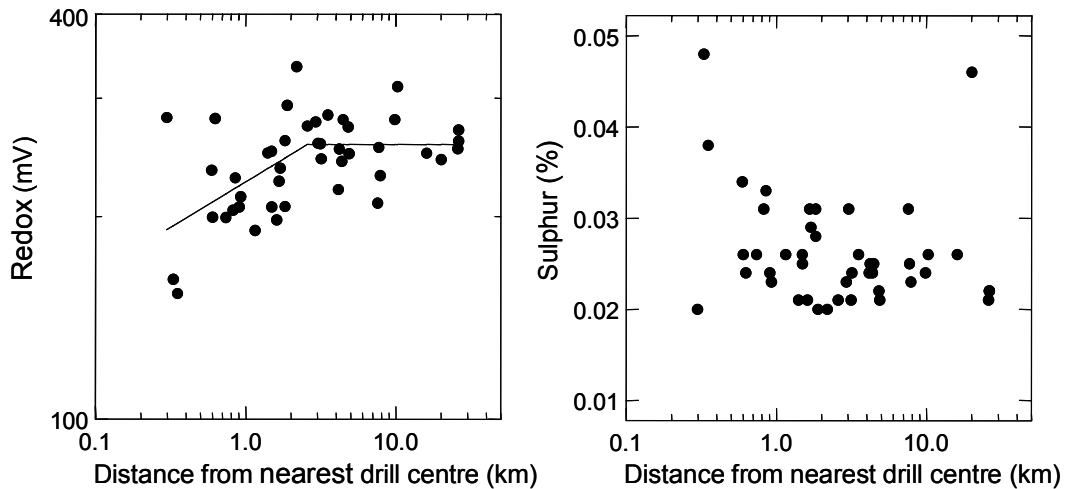




**Figure 5-12 Spatial Distribution of Ammonia (2005)**

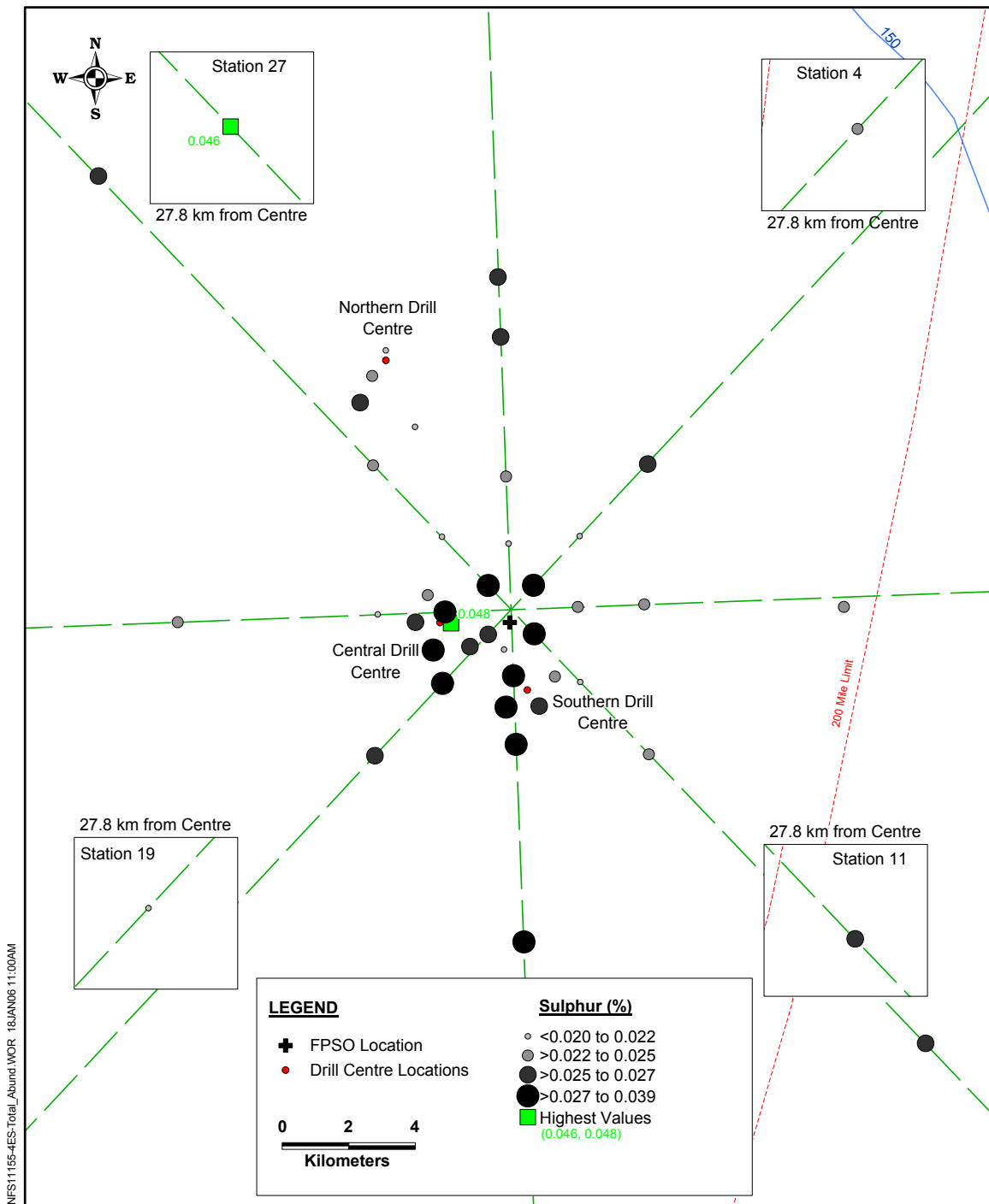
Note: - Highest values were identified as outliers by SPSS 14 software

Over all stations, sulphur concentrations were uncorrelated with depth or distance (Table 5-10). Concentrations were higher near the Central and Southern drill centres than at most other stations, but were also high at station 27 (Northwest reference station) and low near the Northern drill centre (Figures 5-13 and 5-14). Sulphur is a good example of a variable that was significantly although weakly correlated with tracer concentrations (Table 5-8), *may* be affected by drilling in the immediate vicinity of some drill centres, but with extreme values also occurring at more remote stations.



**Figure 5-13 Sulphur and Redox versus Distance from the Nearest Drill Centre (2005)**

Redox levels increased with distances from drill centres and were uncorrelated with depth (Table 5-10; Figure 5-13). Redox levels were lowest near the Central and Southern drill centres, although any reductions attributable to drilling appear localized (Figure 5-15). Redox levels near the Northern drill centre were similar to or higher than levels elsewhere (note the negative and significant partial  $r$  for  $N d$  in Table 5-10). Thus, the spatial distribution of redox was unique, with evidence of potential adverse effects (reductions) from the Central and Southern drill centres and potential positive effects (increases) from the Northern drill centre. Fitting a hockey-stick model based on distance from the nearest drill centre significantly reduced error variances relative to a bivariate log-log regression on distance from the nearest drill centre (Table 5-11; Figure 5-13). The estimated zone of influence was 2.6 km. However, given the difference in direction or sign of effects among drill centres, a model with distances from each drill centre as  $X$  variables was an even better fit (depth can be ignored) (Table 5-11). The parametric full distance model in Table 5-12 indicates that reductions in redox near the Central drill centre were greater and more significant than any increases near the Northern drill centre, a reasonable conclusion based on the spatial distribution in Figure 5-15.



**Figure 5-14 Spatial Distribution of Sulphur (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software

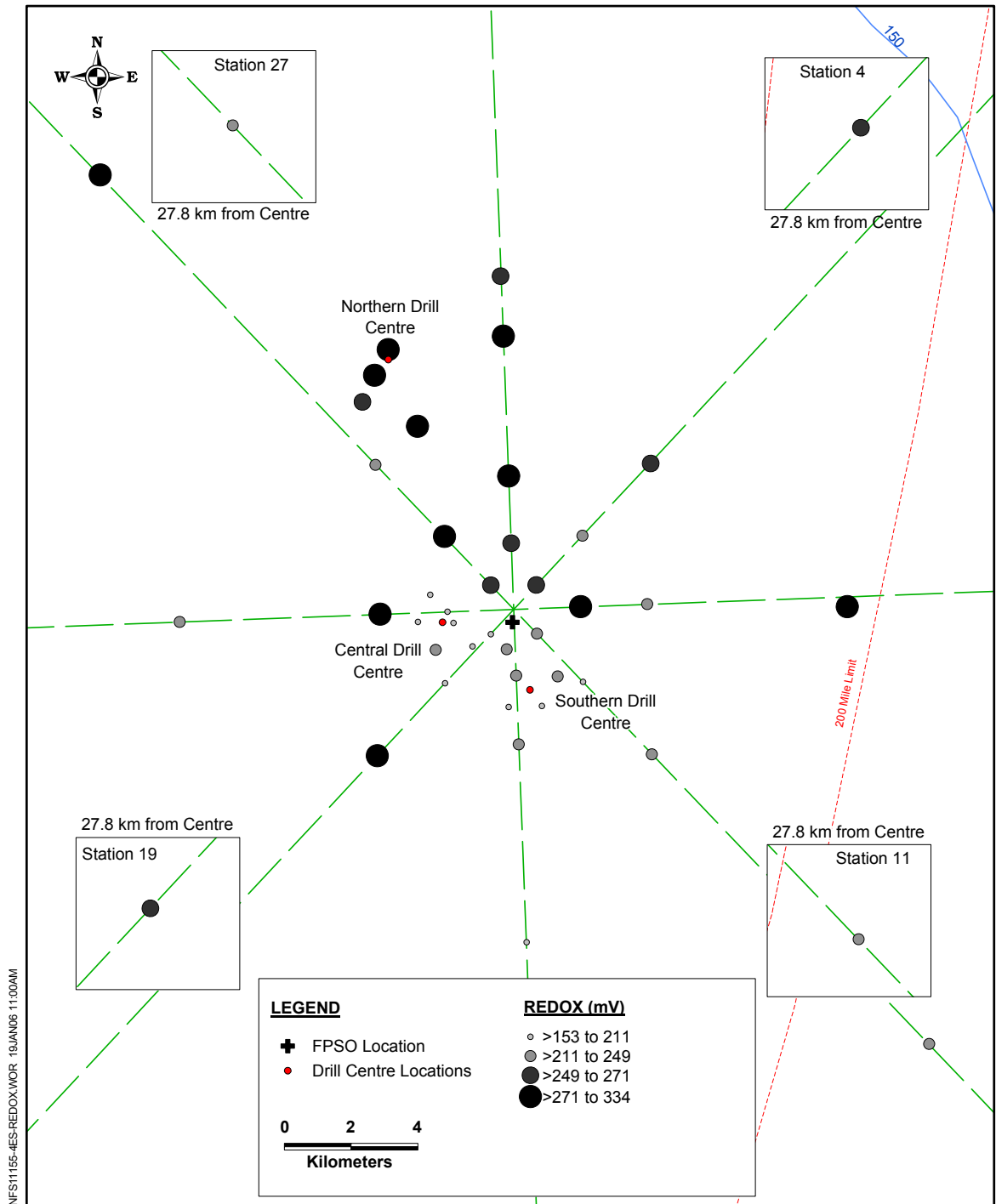


Figure 5-15 Spatial Distribution of Redox (2005)

**Table 5-12 Results of Regression of Redox Levels on Distances from the Three Drill Centres (2005)**

Term/Test	Result	Value
Overall model	<i>R</i>	<b>0.701***</b>
All drill centres versus nearest	$\rho$	<b>&lt;0.001</b>
Constant	Intercept ( <i>a</i> )	2.368 ± 0.023
Distance from:		
Northern drill centre	Slope ( <i>b</i> )	-0.061 ± 0.021**
Central drill centre	Slope ( <i>b</i> )	<b>0.085 ± 0.023***</b>
Southern drill centre	Slope ( <i>b</i> )	0.017 ± 0.024

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - All variables were log-transformed

### 5.4.1.3 Comparison Among Years (2000, 2004, 2005)

Table 5-13 provides results of RM regression models comparing sediment physical and chemical characteristics among the three sample years (2000, 2004, 2005) for the 37 stations sampled in all three years. Table 5-14 provides results of RM regression analyses comparing the two EEM years (2004, 2005) for the 42 stations sampled in both years; stations 4 and 19 with extreme depth values were excluded. In 2000, ammonia and sulphur were not measured and all >C<sub>10</sub>-C<sub>21</sub> HC concentrations were below EQL. For interpretation of results (also see B-5 for further details):

- The Among Stations terms test for relationships between Y variables and depth or distance common to all years (i.e., relationships between mean Y and X). The Among Stations Error 1 term tests for *carry-over* effects, or persistent differences among stations unrelated to depth or distance.
- The Within Stations Year terms test for differences among years common to all or most stations. The Within Stations Year × X terms test for changes in slopes of Y versus X relationships among years (significant effects of X on differences in Y among years). When changes in X effects (i.e., significant Year × X effects) occur, overall or Among Stations X effects should be interpreted with caution (i.e., differences are usually more important than averages).
- For the analysis of all three years, Within Stations differences among years can be divided into differences or *contrasts* between 2000 versus 2004 and 2005 (baseline versus EEM years), and between 2004 versus 2005. If drilling effects occurred, slopes of distance relationships should change after drill centres became active (i.e., Year × *d* terms should be significant for before versus after drilling).
- For the analysis of the two EEM years (2004, 2005), Within Stations results should be similar to Within Stations results for the 2004 versus 2005 contrast for the three-year data set (i.e., adding five stations should not result in large changes). The two-year Among Stations results may reflect a mix of natural and project-related effects common to both EEM years.
- Both the three- and two-year data sets provided strong tests of effects from the Central and Southern drill centres, but weak tests of effects from the Northern drill centre. The set of stations re-sampled over time was biased towards the centre of

the White Rose development and locations closer to the Central and Southern drill centres.

- Results are expressed as *F* values, which are estimates of effect sizes. *F* values greater than 1 indicate added variance attributable to the terms tested.

**Table 5-13 Results of RM Regression Analysis Comparing Sediment Physical and Chemical Characteristics Among 2000, 2004 and 2005**

Term	df	F value for Y variable				
		Barium	% fines	TOC	Metals PC1	Aluminum
<i>Among Stations</i>						
Depth	1,32	0.02	<b>14.52***</b>	1.02	0.86	2.29
Northern (N) <i>d</i>	1,32	4.11	0.04	0.57	0.85	0.99
Central (C) <i>d</i>	1,32	0.00	1.24	<b>15.00***</b>	0.15	0.10
Southern (S) <i>d</i>	1,32	<b>51.10***</b>	5.34*	2.51	3.45	3.82
Error 1 <sup>1</sup>	32,64	1.60	1.60	1.52	1.09	1.67*
<i>Within Stations</i>						
<i>Overall</i>						
Year	2,64	0.02	1.35	0.79	0.83	0.04
Year × Depth	2,64	0.04	2.27	0.79	0.85	0.08
Year × N <i>d</i>	2,64	0.06	3.06	1.34	0.69	0.58
Year × C <i>d</i>	2,64	<b>20.54***</b>	0.46	0.47	0.60	0.21
Year × S <i>d</i>	2,64	<b>30.79***</b>	0.18	1.08	2.26	1.42
<i>2000 versus 2004-05</i>						
Year	1,32	0.03	0.32	0.67	0.43	0.07
Year × Depth	1,32	0.08	0.60	0.71	0.40	0.14
Year × N <i>d</i>	1,32	0.09	1.44	1.73	1.15	0.20
Year × C <i>d</i>	1,32	1.67	1.46	0.07	0.16	0.46
Year × S <i>d</i>	1,32	<b>43.59***</b>	2.76	0.68	0.66	1.66
<i>2004 versus 2005</i>						
Year	1,32	0.02	2.46	0.92	1.25	0.02
Year × Depth	1,32	0.00	4.06	0.88	1.31	0.04
Year × N <i>d</i>	1,32	0.04	4.78*	0.87	0.21	0.82
Year × C <i>d</i>	1,32	<b>36.92***</b>	0.05	0.94	1.05	0.06
Year × S <i>d</i>	1,32	<b>19.67***</b>	0.74	1.56	3.90	1.27

- Notes:
- Appendix B-5 explains terms and tests in the RM regression model
  - df = degrees of freedom for the numerator (effect) and denominator (error) for *F*
  - *d* = distances from various drill centres
  - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)
  - *n* = 37 stations sampled in all three years
  - Distances and all Y variables except Metals PC1 were log-transformed
  - <sup>1</sup>—Error 1 = carry-over effects or persistent differences among stations unrelated to depth or distance

**Table 5-14 Results of RM Regression Analysis Comparing Sediment Physical and Chemical Characteristics Between 2004 and 2005**

Term	df	F value for Y variable							
		Barium	>C <sub>10</sub> -C <sub>21</sub> HCs	% fines	TOC	Metals PC1	Aluminum	Ammonia	Sulphur
<i>Among Stations</i>									
Depth	1,37	0.00	1.94	11.19**	0.04	0.63	1.23	3.64	0.01
Northern (N) <i>d</i>	1,37	2.44	9.01**	0.00	0.57	0.03	2.09	2.08	0.09
Central (C) <i>d</i>	1,37	0.18	0.00	0.33	4.42*	0.09	0.97	8.76**	1.10
Southern (S) <i>d</i>	1,37	<b>31.94***</b>	<b>42.37***</b>	5.01*	4.37*	5.27*	6.79*	<b>18.32***</b>	0.75
Error 1 <sup>1</sup>	37,37	<b>4.84***</b>	<b>5.37***</b>	<b>3.64***</b>	<b>3.07***</b>	<b>2.83***</b>	<b>2.49***</b>	1.43	<b>3.05***</b>
<i>Within Stations</i>									
Year	1,37	0.01	0.19	2.31	1.16	0.10	0.16	1.81	3.64
Year × Depth	1,37	0.01	0.16	2.51	1.20	0.10	0.16	1.84	5.04*
Year × N <i>d</i>	1,37	0.91	4.46*	4.03	0.02	0.22	0.17	0.50	<b>13.49***</b>
Year × C <i>d</i>	1,37	<b>61.79***</b>	<b>68.77***</b>	1.60	1.67	1.34	0.02	2.40	2.31
Year × S <i>d</i>	1,37	<b>29.90***</b>	<b>14.83***</b>	0.90	2.40	4.34*	1.27	5.66*	6.17*

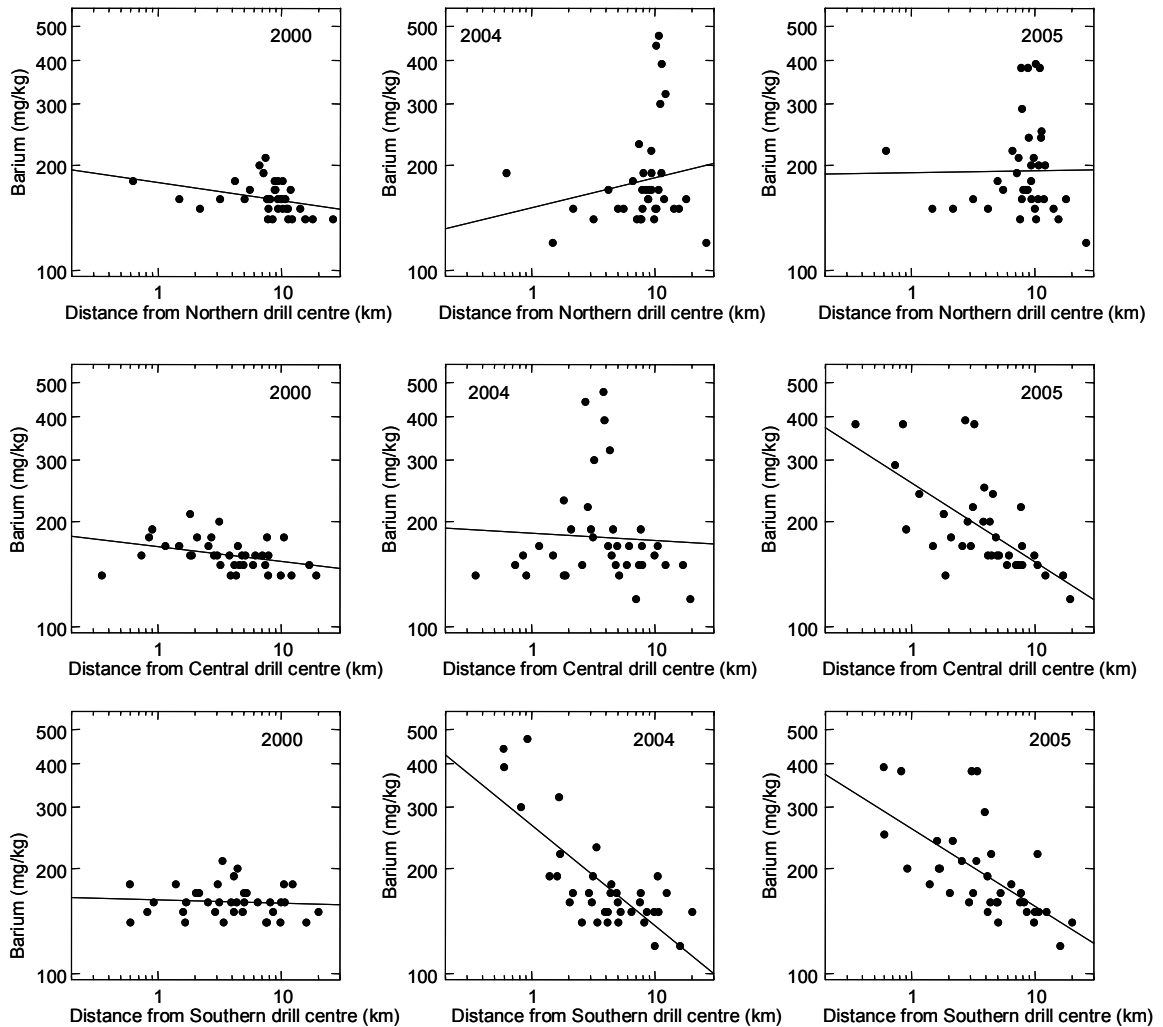
Notes:

- Appendix B-5 explains terms and tests in the RM regression model
- df = degrees of freedom for the numerator (effect) and denominator (error) for *F*
- *d* = distances from various drill centres
- \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)
- $n = 42$  stations sampled in both years
- Distances and all Y variables except Metals PC1 were log-transformed
- <sup>1</sup>—Error 1 = carry-over effects or persistent differences among stations unrelated to depth or distance

### Tracers

Results for barium and >C<sub>10</sub>-C<sub>21</sub> HCs provided clear evidence of effects of drilling at the Central and Southern drill centres on concentrations of the two tracers. For barium, relationships with distance from the Northern drill centre did not vary over time (i.e., Year × Northern *d* terms were not significant in Tables 5-13 and 5-14) (Figure 5-16). Concentrations greater than 250 mg/kg have never been observed at stations within 2 km of the Northern drill centre, even for stations sampled only in 2004 (Husky Energy 2005) and excluded from the RM analysis and Figure 5-16. In contrast, the relationship with distance from the Central drill centre changed significantly and substantially between 2004 and 2005 after drilling began (Tables 5-13 and 5-14; Figure 5-16).

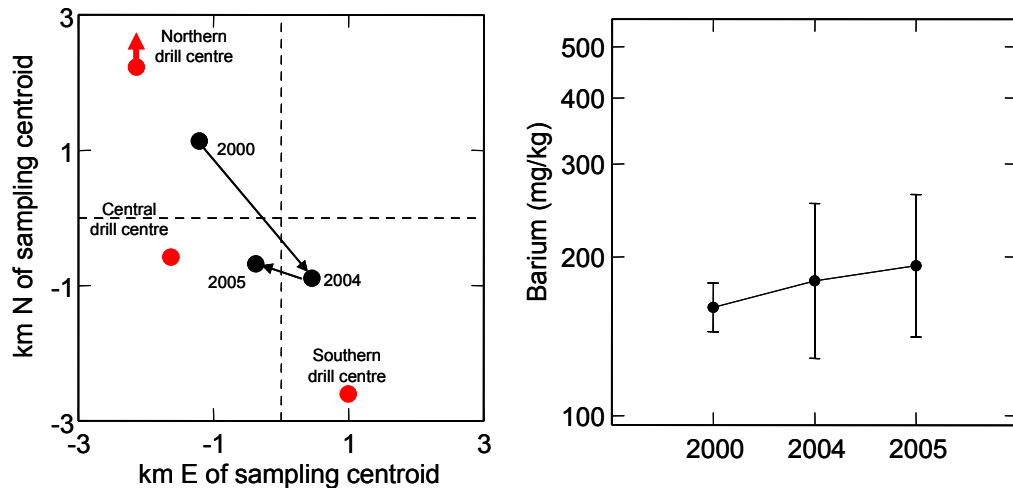
Relationships between barium and distance from the Southern drill centre also changed significantly and substantially between 2000 and 2004 after drilling began (Figure 5-16). The distance gradient was weaker in 2005 than in 2004 because barium levels decreased at stations within 2 km of the Southern drill centre. The change in distance relationships between 2004 and 2005 was highly significant ( $p \leq 0.001$  for Year × Southern *d* term for 2004 versus 2005 in Table 5-13, and for the comparison of the two years in Table 5-14). The change between 2004 and 2005 appears small in Figure 5-16, but bivariate plots of Y versus a single X variable can conceal or inflate the effects of that X variable because the effects of other X variables are ignored.



**Figure 5-16 Barium Concentrations versus Distances from the Three Drill Centres for 37 Stations Sampled in 2000, 2004 and 2005**

Figure 5-17 provides barium centroids (left plot), and overall changes in barium concentrations over time (right plot), for the 37 stations sampled in all three years. The sample design and sampling centroid (North and East coordinates=0,0) were biased towards the Central and Southern drill centres. In 2000, the barium centroid was to the Northwest of the sampling centroid, but moved southeast towards the Southern drill centre in 2004 (despite drilling at the Northern drill centre), and then east towards the Central drill centre in 2005 after drilling began at that centre.



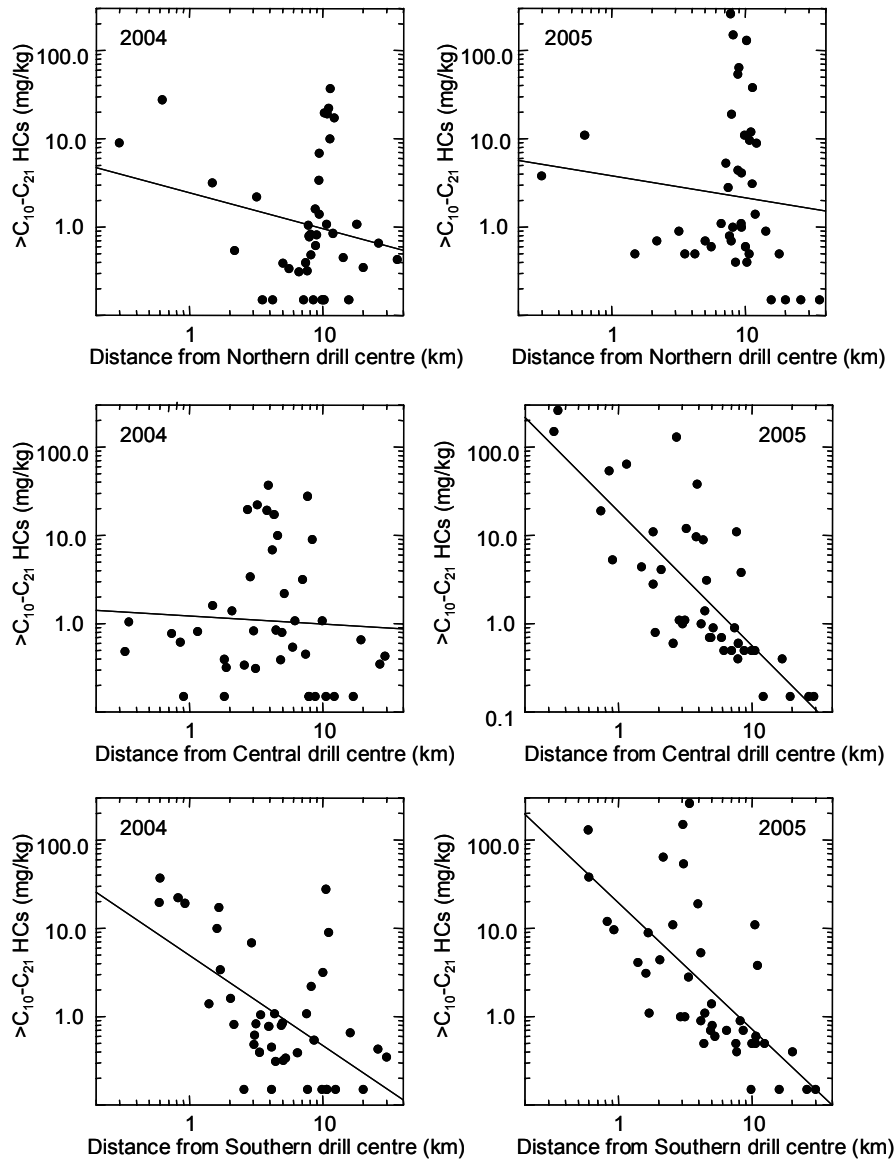


**Figure 5-17 Barium Centroids and Changes in Concentrations Over Time for 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used (e.g., logarithmic for barium). The Y axes include the full range of individual values

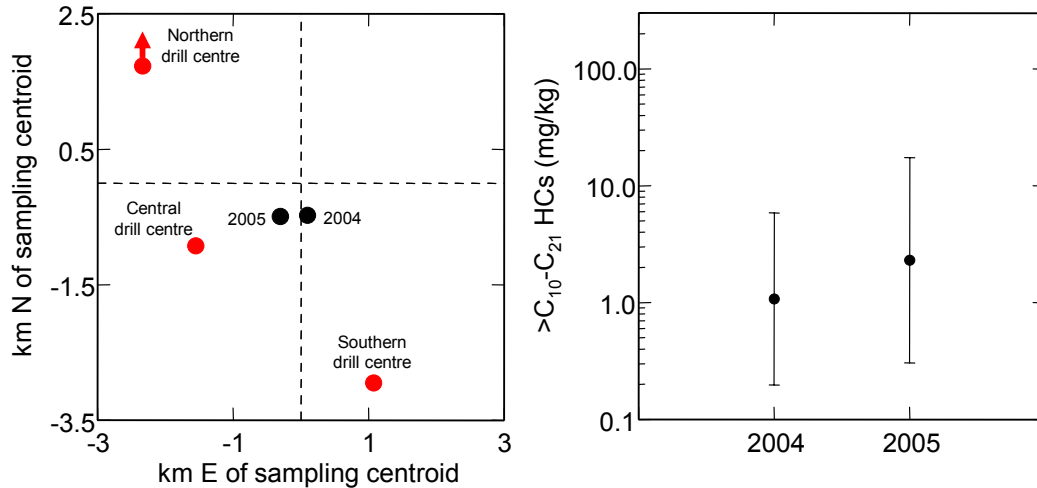
The net result was that barium concentrations over the 37 stations increased over time (Figure 5-17). Most of the increase was attributable to increased concentrations near active drill centres. Barium concentrations at more remote stations, far from the two drill centres, did not increase over time in Figure 5-17, and Year terms testing for changes common to all or most were not significant in the RM regression analyses (Tables 5-13 and 5-14).

With all  $>C_{10-C_{21}}$  HC concentrations in 2000 less than EQL, baseline distance relationships would be horizontal lines. In 2004 and 2005,  $>C_{10-C_{21}}$  HC concentrations decreased with distance from the Northern and Southern drill centres and, in 2005, decreased with distance from the Central drill centre (Figure 5-18). The distance gradients for the Northern and Southern drill centres over the two EEM years were significant (Among Stations distance terms in Table 5-14), with gradients stronger and more significant for the Southern drill centre. Distance gradients for the Northern and Southern drill centres also changed significantly between 2004 and 2005 (Year  $\times d$  terms in Table 5-14). Between 2004 and 2005,  $>C_{10-C_{21}}$  HC concentrations within 2 km of the Northern drill centre decreased and concentrations within 2 to 3 km of the Southern drill centre increased (Figure 5-18). These changes between 2004 and 2005 appear small in the bivariate plots in Figure 5-18 because they are obscured by changes in effects from the Central drill centre at intermediate distances. The change in distance gradients between 2004 and 2005 (i.e., after drilling began) for the Central drill centre were highly significant (Year  $\times$  Central  $d$  term in Table 5-14), with concentrations within 2 to 3 km increasing 10- to 100-fold (Figure 5-18). Finally, depth relationships over both EEM years combined, and any changes in these relationships, were not significant for  $>C_{10-C_{21}}$  HCs, despite apparent depth effects for all stations sampled in 2005 (Section 5.4.1.2).



**Figure 5-18 >C<sub>10</sub>-C<sub>21</sub> HC Concentrations versus Distances from the Three Drill Centres for 42 Stations Sampled in 2004 and 2005**

Figure 5-19 provides centroids and overall changes over time for >C<sub>10</sub>-C<sub>21</sub> HCs. In 2000, the >C<sub>10</sub>-C<sub>21</sub> HC centroid would be at the sampling centroid (0,0). >C<sub>10</sub>-C<sub>21</sub> HC centroids in 2004 and 2005 were also close to the sampling centroid, illustrating one limitation of the use of centroids. Centroids for variables unaffected by distance or direction (i.e., varying randomly) will be close to the sampling centroid, but so will centroids for variables affected by all three drill centres or just the Northern and Southern drill centres (e.g., >C<sub>10</sub>-C<sub>21</sub> HCs in 2004 and 2005). The Northern drill centre had some effect on >C<sub>10</sub>-C<sub>21</sub> HC concentrations. If this were not true, the 2004 centroid would be further southeast and closer to the Southern drill centre, and the 2005 centroid further to the southwest and between the Central and Southern drill centres.



**Figure 5-19 >C<sub>10</sub>-C<sub>21</sub> HC Centroids and Changes in Concentrations Over Time for 42 Stations Sampled in 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

In 2004, overall >C<sub>10</sub>-C<sub>21</sub> HC concentrations increased to approximately 1 mg/kg versus less than 0.3 mg/kg at all stations in 2000 (Figure 5-19; 2000 concentrations would be at or near the bottom of the Y axis). In 2005, there was a further increase to approximately 2 mg/kg (i.e., overall concentrations doubled). These increases are entirely attributable to drilling.

For 2004, as for 2005 (Section 5.4.1.2), hockey-stick or threshold distance regression models for both tracers and all stations sampled significantly ( $p < 0.001$ ) reduced error variances relative to bivariate log-log regressions (Table 5-15). The distance measure used for 2004 was distance to the nearest of the two active drill centres (Northern and Southern). For both variables, estimated background or blade concentrations were similar between years, suggesting that natural changes were minimal. For barium, the slope of the shaft for 2004 was steeper and the estimated zone of influence smaller (2.4 versus 3.6 km, although CIs overlapped considerably) for 2004 than for 2005, suggesting that the spatial extent of contamination increased in 2005. For >C<sub>10</sub>-C<sub>21</sub> HCs, regression slopes and intercepts were similar for the two years. Estimated zones of influence were 5.3 km in 2004 and 6.5 km in 2005. These values should be considered similar, given the wide CIs. In both years, concentrations decreased by at least 1,000-fold with distance up to 10 km from drill centres. These strong distance gradients common to both years overwhelmed any effect that the two-fold difference in overall concentrations (Figure 5-19) had on estimates of the zones of influence.

**Table 5-15 Results of Hockey-stick (Threshold) Regressions on Distance from the Nearest Active Drill Centre for Barium and >C<sub>10</sub>-C<sub>21</sub> HCs (2004 and 2005)**

Result/Estimate	Barium		>C <sub>10</sub> -C <sub>21</sub> HCs	
	2004	2005	2004	2005
Overall <i>R</i>	<b>0.776***</b>	<b>0.767***</b>	<b>0.825***</b>	<b>0.875***</b>
<i>p</i> for adding threshold	<0.001	0.015	<0.001	0.016
antilog <i>a</i> (blade/background value)	156	149	0.31	0.26
95% CI	144 to 169	131 to 169	0.18 to 0.56	0.14 to 0.49
<i>b</i> (distance slope for shaft)	-0.617	-0.383	-1.90	-1.89
95% CI	-0.823 to -0.411	-0.520 to -0.246	-2.30 to -1.50	-2.32 to -1.46
antilog <i>X<sub>T</sub></i> (threshold distance in km)	2.4	3.6	5.3	6.5
95% CI	1.6 to 3.5	2.1 to 6.3	3.7 to 7.8	3.9 to 10.8

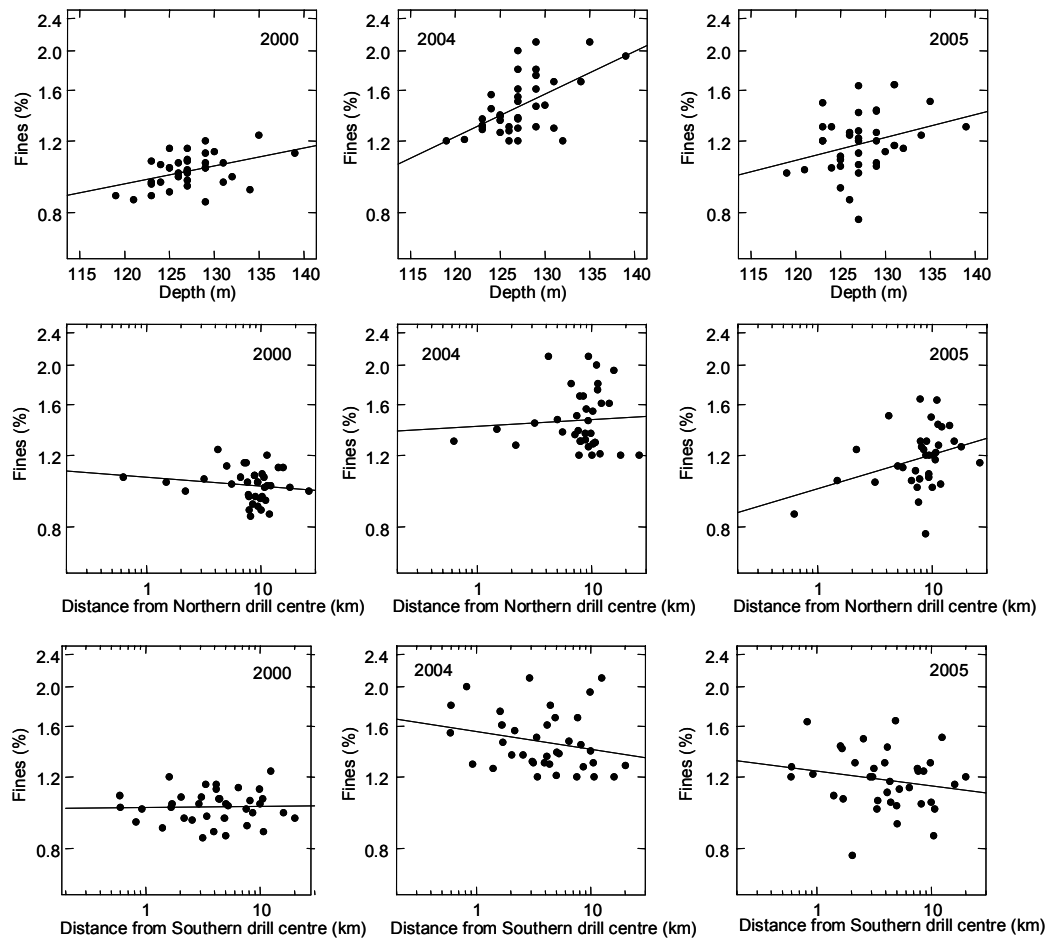
Notes:

- \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)
- *X* was distance from the nearest active drill centre (Northern, Southern in 2004; Northern, Central, Southern in 2005)
- $n = 56$  stations in 2004 and 44 stations in 2005
- All variables were log-transformed

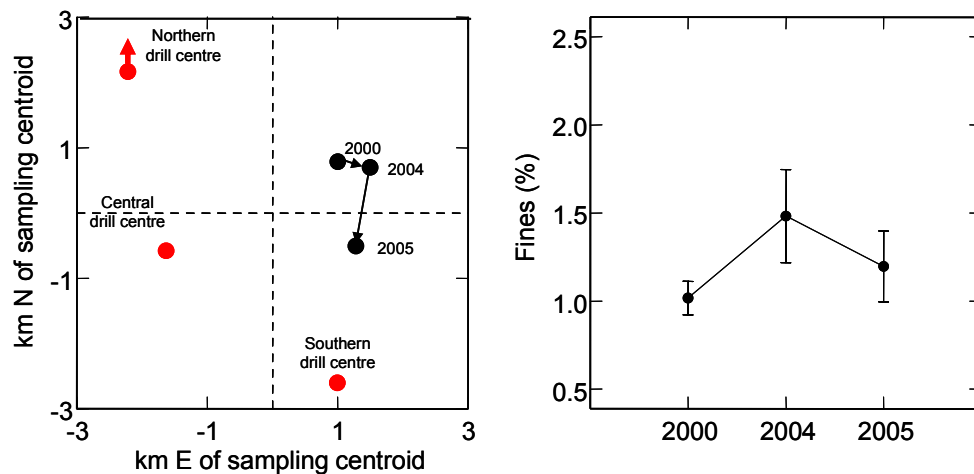
### Fines and TOC

Fines content increased with depth in all three sample years (Figure 5-20). Depth relationships did not differ among years (Year  $\times$  Depth terms in Tables 5-13 and 5-14), and the common depth relationships over time were highly significant (Among Stations Depth terms in Tables 5-13 and 5-14). For the 37 stations sampled in all three years, the relationship between fines content and distance from the Northern drill centre changed significantly from no relationship in 2004 (and 2000) to an increase in fines content with distance in 2005 (Figure 5-20; Year  $\times$  Northern *d* term for the 2004 versus 2005 contrast in Table 5-13). The same change occurred between 2004 and 2005 for the 42 stations sampled in both years but was not quite significant ( $0.05 < p < 0.10$ ). The opposite change appeared to occur with distance from the Southern drill centre (i.e., a change from no relationship in 2000 to a weak decrease with distance in 2004 and 2005) (Figure 5-20). However, differences in these gradients over time were not significant in the three-year RM regression model (Within Stations Year  $\times$  Southern *d* terms in Table 5-13). Instead, there was a weak but significant decrease with distance from the Southern drill centre over all three years combined (Among Stations Southern *d* term in Table 5-13). There was no evidence from either the three- or two-year RM regression models for any relationship between fines content and distance from the Central drill centre nor any change after drilling began there.

Fines content has always been greater to the Northeast, where depths are greater, as the centroids in Figure 5-21 indicate. Fines content increased between 2000 and 2004, and then decreased in 2005. These changes were natural (or methodological), given the absence of any evidence for broad-scale project effects on fines content.



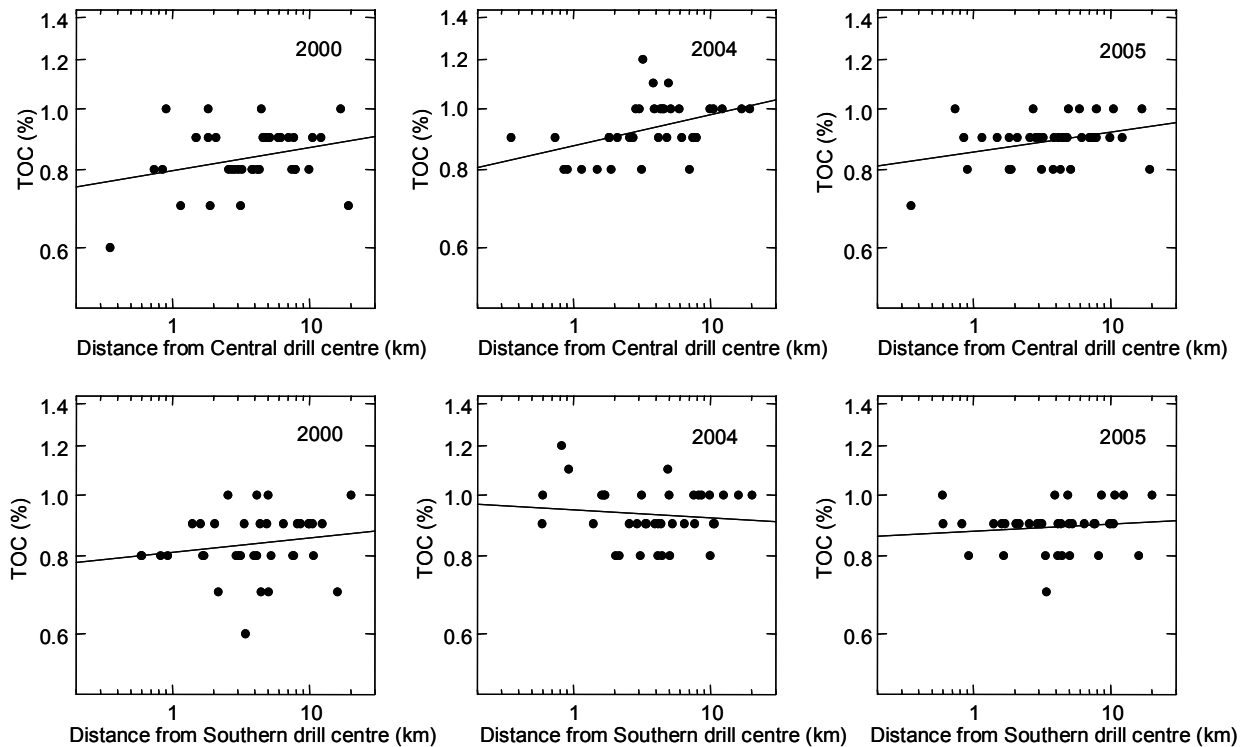
**Figure 5-20 Fines Content versus Depth and Distances from the Northern and Southern Drill Centres for 37 Stations Sampled in 2000, 2004 and 2005**



**Figure 5-21 Fines Centroids and Changes in Fines Content Over Time for 37 Stations Sampled in 2000, 2004 and 2005**

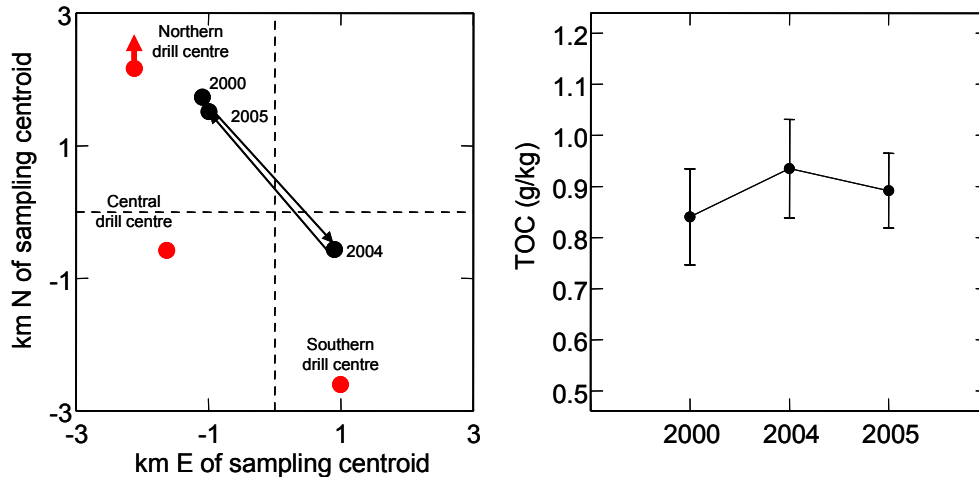
Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

For TOC, there was a consistent and significant increase in TOC with distance from the Central drill centre in all three sample years (Figure 5-22; Among Stations Central *d* terms in Tables 5-13 and 5-14). The cause of this relationship is unknown but presumably natural rather than project-related since it did not vary over time. The two-year analyses also identified a significant relationship between TOC and distance from the Southern drill centre common to 2004 and 2005 (Table 5-14), but this relationship was not significant (Table 5-13) or visually apparent (Figure 5-22) for the three-year data set.



**Figure 5-22 TOC versus Distances from the Central and Southern Drill Centres for 37 Stations Sampled in 2000, 2004 and 2005**

TOC centroids in all three sample years were north or east of the sampling centroid, or relatively far from the Central drill centre (hence the Central *d* effects; Figure 5-23). Centroid locations in 2000 and 2005 were similar, but the 2004 centroid was displaced to the southeast. For TOC, the centroids inflate changes in spatial distribution over time, which were not significant (Within Stations terms in Tables 5-13 and 5-14). TOC concentrations have always varied little within a narrow range between 0.6 to 1.2 g/kg with most values between 0.8 to 1.0 g/kg, as the distance plots in Figure 5-22 and the annual means in Figure 5-23 indicate.



**Figure 5-23 TOC Centroids and Changes in TOC Content Over Time for 37 Stations Sampled in 2000, 2004 and 2005**

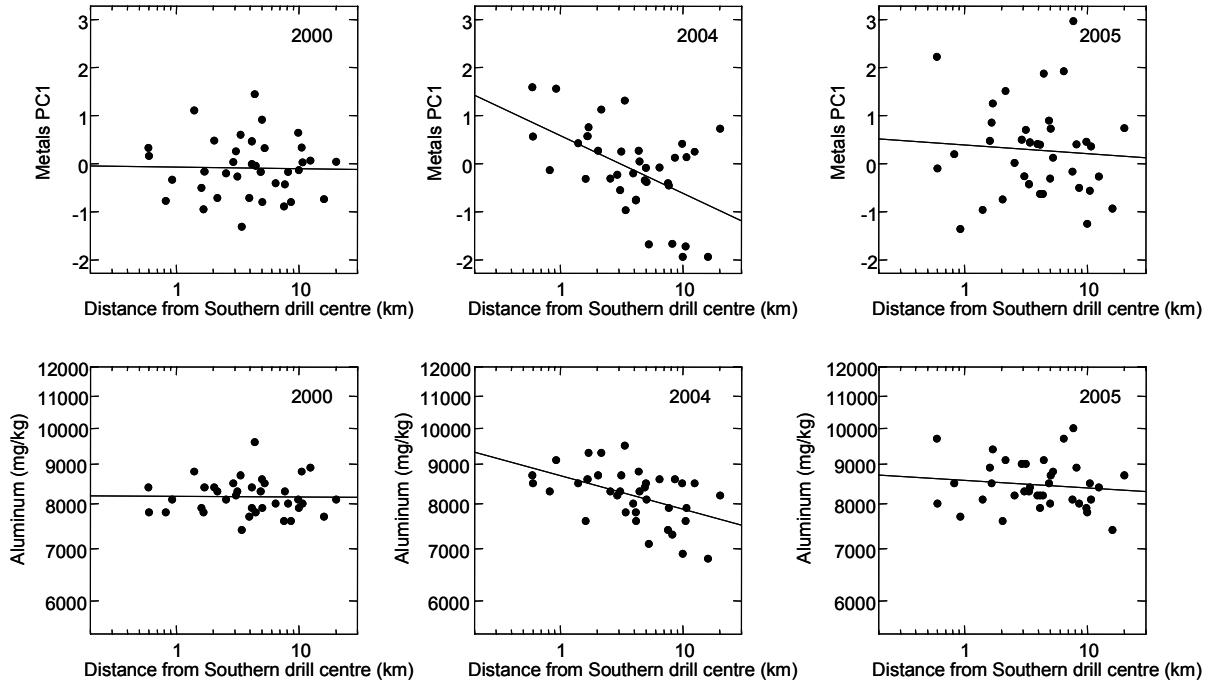
Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

**Metals**

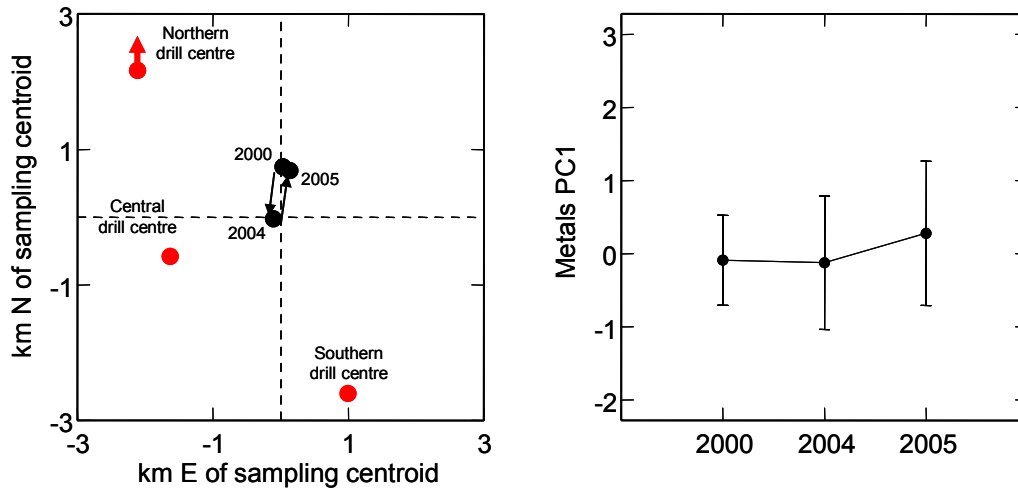
RM regression results for Metals PC1 and aluminum in Tables 5-13 and 5-14 were similar because Metals PC1 was strongly correlated with aluminum concentrations (Table 5-7). For the three-year analyses, no term was significant for either variable, except for Among Stations Error 1 or carry-over effects for aluminum. In 2000, there was no relationship between Metals PC1 and aluminum versus distance from the Southern drill centre; in 2004 both variables decreased with distance; in 2005 the decrease with distance was weaker (Figure 5-24). Changes in the distance gradients, and/or common gradients over time (i.e., Within and Among Stations Southern *d* terms) were not significant, but did provide some larger *F* values, for the three-year analyses (Table 5-13); and changes were sometimes significant for the two-year comparison of 2004 and 2005 (Table 5-14).

Note that any decrease in metal concentrations, and especially Metals PC1 scores, with distance from the Southern drill centre in 2004 and 2005 was partly to mostly a function of decreases in values at remote stations as opposed to increases near the Southern drill centre (Figure 5-24). There was also no evidence that drilling at the other two drill centres had any effects on metal concentrations.

Metals PC1 and aluminum centroids were displaced to the south and towards the Southern drill centre between 2000 and 2004, but returned to baseline centroid locations in 2005 (Figures 5-25 and 5-26). These changes in centroid locations were expected given the greater decreases with distance from the Southern drill centre in 2004 relative to other years (Figure 5-24). However, 2005 centroids moved further from the Central drill centre between 2004 and 2005, after drilling began at that centre. Furthermore, changes over time were negligible, especially for aluminum.



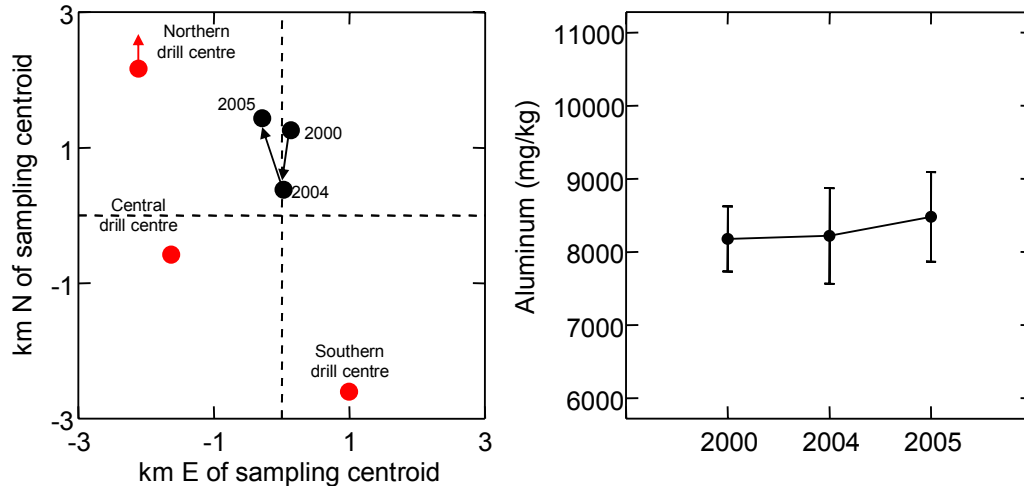
**Figure 5-24 Metals PC1 and Aluminum Concentrations versus Distance from the Southern Drill Centres for 37 Stations Sampled in 2000, 2004 and 2005**



**Figure 5-25 Metals PC1 Centroids and Changes in Values Over Time for 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values





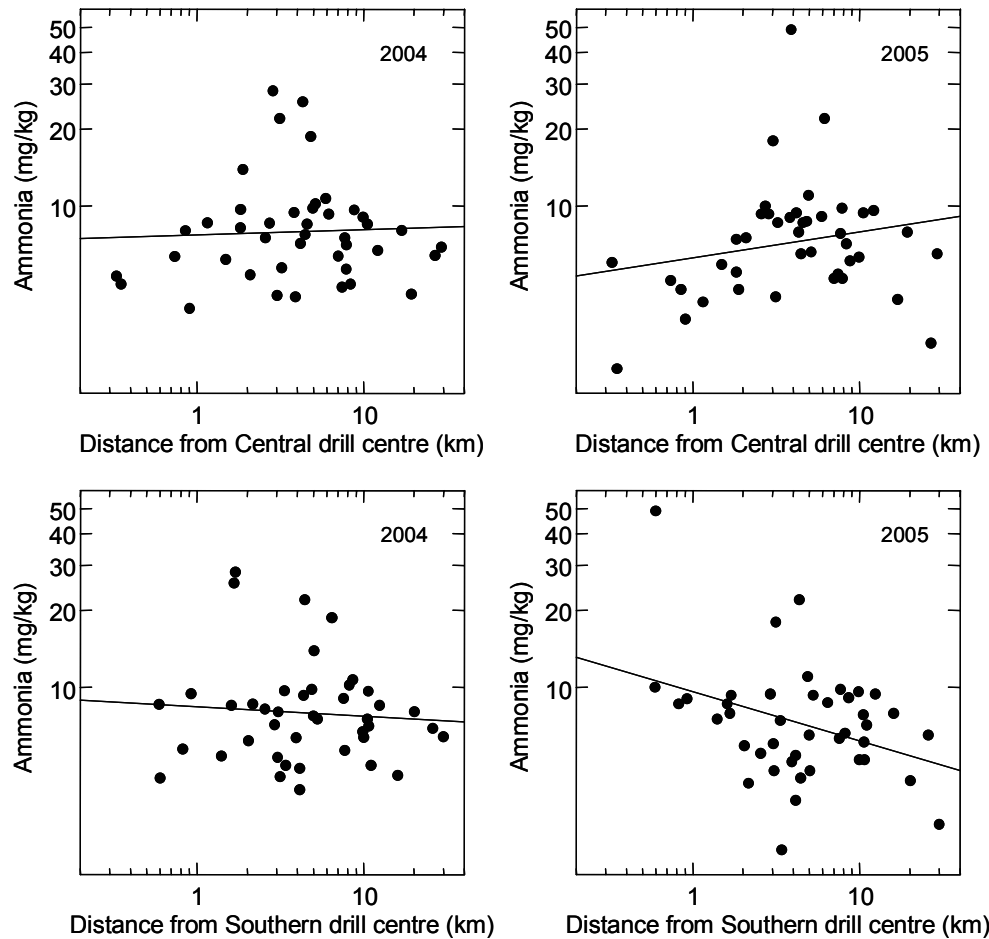
**Figure 5-26 Aluminum Centroids and Changes in Concentrations Over Time for 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

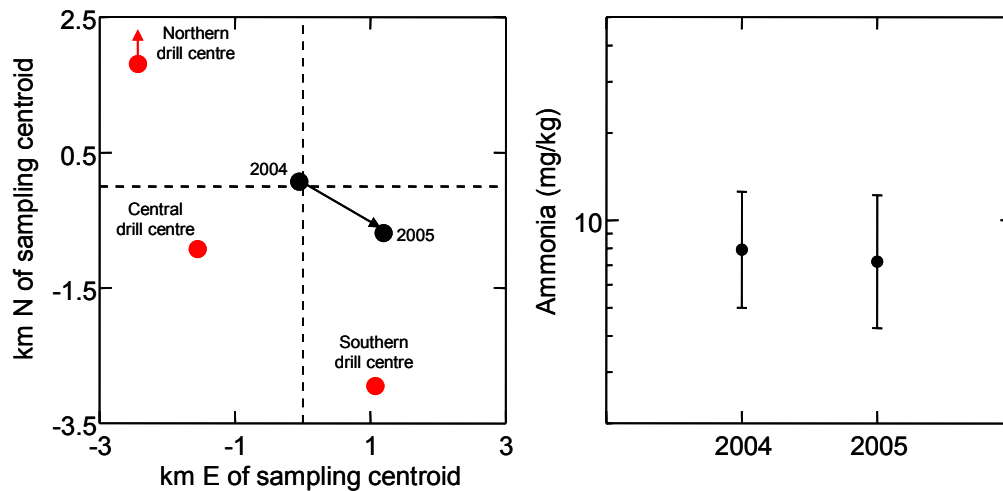
**Ammonia and Sulphur**

Ammonia and sulphur were not measured in 2000, so the only possible comparison was between 2004 and 2005 (Table 5-14). Over both years, there was a significant increase in ammonia concentrations with distance from the Central drill centre, and a significant decrease with distance from the Southern drill centre (Figure 5-27; Among Stations terms in Table 5-14). Both gradients appeared to increase in strength between 2004 and 2005, but only the change in the distance gradient for the Southern drill centre was significant (Within Stations terms in Table 5-14). In 2005, the high ammonia concentration of 49 mg/kg at station S1 was a significant outlier. With this outlier deleted, the distance gradients for the Central and Southern drill centres for both years combined were still significant, but changes in gradients between years were not.

Ammonia centroids moved southeast between 2004 and 2005 (Figure 5-28), partly as a result of the changes in distance gradients for the Central and Southern drill centres shown in Figure 5-27. Over all 42 stations, ammonia concentrations were similar in the two EEM years (Figure 5-28).



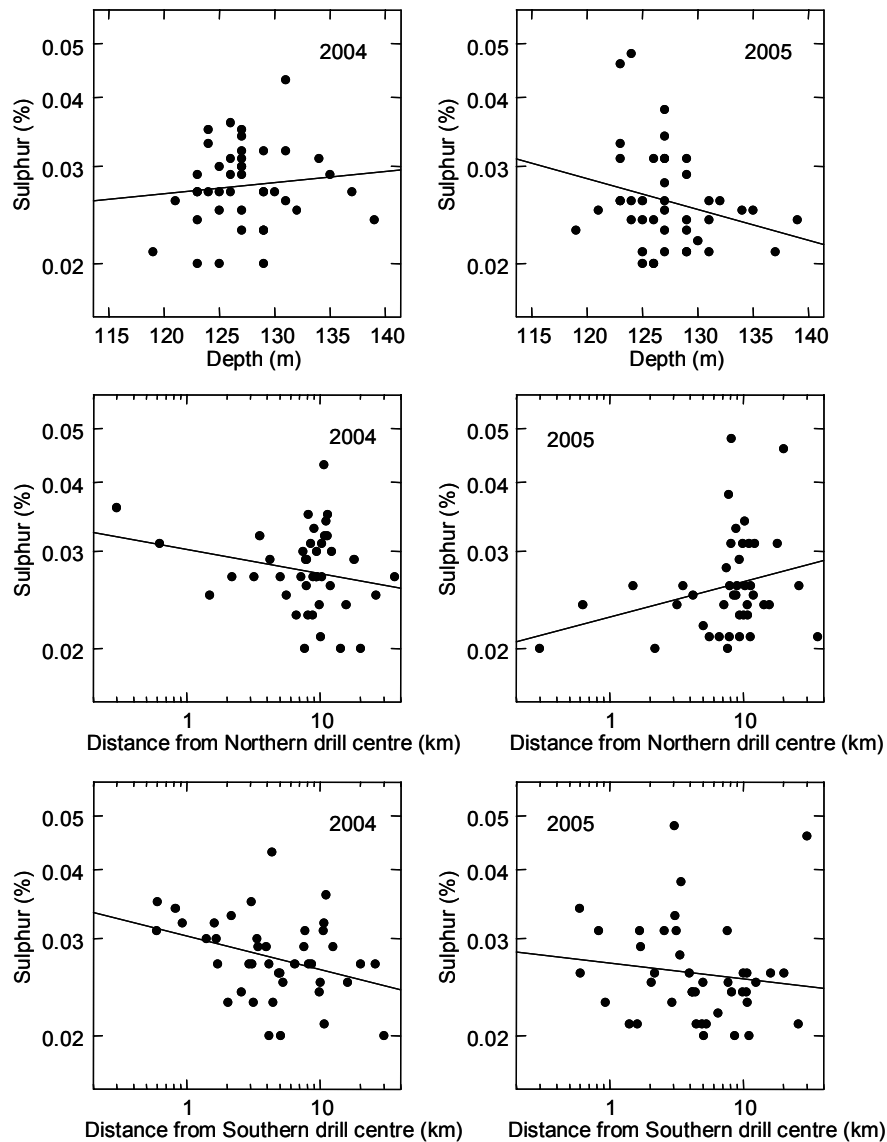
**Figure 5-27 Ammonia Concentrations versus Distance from the Central and Southern Drill Centres for 42 Stations Sampled in 2004 and 2005**



**Figure 5-28 Ammonia Centroids and Changes in Concentrations Over Time for 42 Stations Sampled in 2004 and 2005**

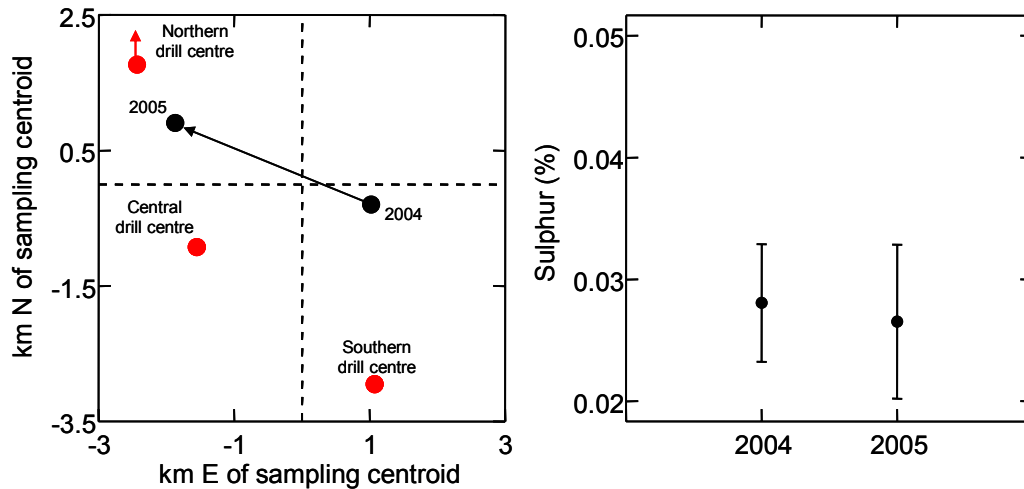
Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

Relationships between sulphur concentrations versus depth and distances from the Northern and Southern drill centres differed significantly between 2004 and 2005 (Within Stations terms in Table 5-14). Relationships with depth and distance from the Northern drill centre were reversed between the two years (Figure 5-29). Relationships with distance from the Southern drill centre appeared similar between years (Figure 5-29), so it was surprising that the change (a weaker gradient in 2005) was significant. It should be noted that there were several outliers relative to the limited variance among most stations for sulphur, and depth and distance regressions were not particularly good fits.



**Figure 5-29 Sulphur Concentrations versus Distance from the Northern and Southern Drill Centres for 42 Stations Sampled in 2004 and 2005**

The sulphur centroid moved substantially north and west between 2004 and 2005 (Figure 5-30) because of the changes in depth and distance gradients. Although spatial distributions changed, overall concentrations did not (Figure 5-30).



**Figure 5-30 Sulphur Centroids and Changes in Concentrations Over Time for 42 Stations Sampled in 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

**Carry-over Effects**

For barium, fines, TOC, Metals PC1 and aluminum, measured in all three years, Within Stations results for the two-year comparison in Table 5-14 were similar to Within Stations results for the 2000 versus 2004 and 2005 contrast in the three-year comparison in Table 5-13, as expected. However, for all five variables, Error 1 or carry-over effects were much stronger and more significant for the two-year comparison than for the three-year comparison. Carry-over effects between 2004 and 2005 were also significant for >C<sub>10</sub>-C<sub>21</sub> HCs and sulphur, but not ammonia (Table 5-14). For barium and >C<sub>10</sub>-C<sub>21</sub> HCs, carry-over effects or persistent differences from 2004 to 2005 may be largely attributable to project-related directional and other effects unrelated to distance. However, there was little or no evidence of project effects on other variables. Instead, natural carry-over effects may be important over the short term (one or two years) but not over the longer term (three or more years).

**5.4.2 Toxicity**

In 2005, as in 2004 and 2000 (Husky Energy), all Microtox IC50s were >197,000 mg/kg (the highest concentration tested), indicating that there were no toxic effects on luminescent bacteria. Analysis results for 2005 are provided in Appendix B-6.

Amphipod toxicity was not noted in 2000 and 2004. In 2005, sediment from one station was toxic to amphipods (survival: 28%), and survival in sediment from another station (67.5%) was lower than in samples from other stations sampled in 2000, 2004 and 2005 (survival has usually been greater than 80%) (Table 5-16 and Appendix B-7).

**Table 5-16 Amphipod Toxicity Trials Summary Data and Interpretation**

Distance (km) to Nearest Drill Centre	Station	Year	Amphipod Survival (%)	Sample Standard Deviation	Comparison to Laboratory Controls				Comparison to Reference Stations			
					Dunnett's t-stat	Statistically Significant	≥ 30% Reduction in Survival	Interpretation (Toxic / Nontoxic)	Dunnett's t-stat	Statistically Significant	≥ 20% Reduction in Survival	Interpretation (Toxic / Nontoxic)
0.3	N4	2000	NS									
		2004	89	6.29	2.675	Yes	No	Nontoxic	0.931	No	No	Nontoxic
		2005	93	8.2	-1.49	No	No	Nontoxic	1.463	No	No	Nontoxic
0.31	S5	2000	NS									
		2004	74	4.79	7.033	Yes	No	Nontoxic	4.645	No	No	Nontoxic
		2005	NS				No	No				
0.33	C5	2000	NS									
		2004	85	4.08	3.969	Yes	No	Nontoxic	1.945	No	No	Nontoxic
		2005	94	5.8	0.84	No	No	Nontoxic	2.716	No	No	Nontoxic
0.35	20	2000	97	2.7	0.3	No	No	Nontoxic				
		2004	94	4.79	1.654	No	No	Nontoxic	0.565	No	No	Nontoxic
		2005	92	9.3	1.11	No	No	Nontoxic	2.039	No	No	Nontoxic
0.59	13	2000	93	5.7	0.23	No	No	Nontoxic				
		2004	88	5	3.349	Yes	No	Nontoxic	1.543	No	No	Nontoxic
		2005	95	5.5	-2.49	No	No	Nontoxic	2.576	No	No	Nontoxic
0.6	S1	2000	92	5.7	1.81	No	No	Nontoxic				
		2004	79	4.79	4.85	Yes	No	Nontoxic	3.584	Yes	No	Nontoxic
		2005	83	2.1	0.57	No	No	Nontoxic	0.71	No	No	Nontoxic
0.63	N3	2000	90	5	1.92	No	No	Nontoxic				
		2004	90	9.35	2.527	Yes	No	Nontoxic	0.443	No	No	Nontoxic
		2005	<b>67.5</b>	39.1	0.95	No	No	Nontoxic	2.527	No	No	Nontoxic
0.74	C3	2000	81	8.9	2.57	No	No	Nontoxic				
		2004	95	5.77	1.34	No	No	Nontoxic	0.969	No	No	Nontoxic
		2005	98	2.7	-0.3	No	No	Nontoxic	4.176	Yes	No	Nontoxic
0.82	S2	2000	94	4.2	1.21	No	No	Nontoxic				
		2004	89	6.29	3.014	Yes	No	Nontoxic	1.016	No	No	Nontoxic
		2005	89	7.4	-0.72	No	No	Nontoxic	0.649	No	No	Nontoxic
0.85	C2	2000	96	4.2	0.67	No	No	Nontoxic				
		2004	86	7.5	3.638	Yes	No	Nontoxic	1.511	No	No	Nontoxic
		2005	93	4.1	1.37	No	No	Nontoxic	2.742	Yes	No	Nontoxic
0.9	C4	2000	93	9.8	0.61	No	No	Nontoxic				
		2004	91	4.79	3.344	Yes	No	Nontoxic	0.269	No	No	Nontoxic
		2005	89	7.4	1.87	No	No	Nontoxic	0.961	No	No	Nontoxic
0.92	S4	2000	88	7.6	1.4	No	No	Nontoxic				
		2004	93	6.45	1.712	No	No	Nontoxic	0.213	No	No	Nontoxic
		2005	91	9.7	-1.17	No	No	Nontoxic	1.783	No	No	Nontoxic
1.1	31	2000	NS									
		2004	89	7.42	2.811	Yes	No	Nontoxic	0.784	No	No	Nontoxic
		2005	91	5.8	-1.28	No	No	Nontoxic	0.946	No	No	Nontoxic
1.15	C1	2000	87	8.3	1.63	No	No	Nontoxic				
		2004	91	4.79	2.315	No	No	Nontoxic	0.249	No	No	Nontoxic
		2005	97	4.1	0	No	No	Nontoxic	4.343	Yes	No	Nontoxic
1.4	S3	2000	91	6.5	0.7	No	No	Nontoxic				
		2004	89	6.29	2.976	Yes	No	Nontoxic	1,016	No	No	Nontoxic
		2005	86	5.4	-0.04	No	No	Nontoxic	0.137	No	No	Nontoxic

Distance (km) to Nearest Drill Centre	Station	Year	Amphipod Survival (%)	Sample Standard Deviation	Comparison to Laboratory Controls				Comparison to Reference Stations			
					Dunnett's t-stat	Statistically Significant	≥ 30% Reduction in Survival	Interpretation (Toxic / Nontoxic)	Dunnett's t-stat	Statistically Significant	≥ 20% Reduction in Survival	Interpretation (Toxic / Nontoxic)
1.49	N2	2000	91	8.9	2.89	Yes	No	Nontoxic				
		2004	98	5	0.529	No	No	Nontoxic	1.825	No	No	Nontoxic
		2005	94	3.8	0.99	No	No	Nontoxic	3.089	Yes	No	Nontoxic
1.49	16	2000	92	7.6	1.44	No	No	Nontoxic				
		2004	84	2.5	3.709	Yes	No	Nontoxic	2.627	No	No	Nontoxic
		2005	93	5.2	1.2	No	No	Nontoxic	2.821	Yes	No	Nontoxic
1.61	9	2000	94	6.5	1.21	No	No	Nontoxic				
		2004	94	4.79	0.953	No	No	Nontoxic	0.487	No	No	Nontoxic
		2005	<b>28</b>	21.9	3.37	Yes	Yes	<b>Toxic</b>	11.44	Yes	Yes	<b>Toxic</b>
1.67	14	2000	97	4.5	0.26	No	No	Nontoxic				
		2004	91	8.54	1.997	No	No	Nontoxic	0.087	No	No	Nontoxic
		2005	83	4.2	0.87	No	No	Nontoxic	0.967	No	No	Nontoxic
1.7	8	2000	86	8.2	1.87	No	No	Nontoxic				
		2004	89	8.54	1.88	No	No	Nontoxic	0.750	No	No	Nontoxic
		2005	88	6.8	-0.76	No	No	Nontoxic	0.573	No	No	Nontoxic
1.83	17	2000	91	8.9	1.68	No	No	Nontoxic				
		2004	91	6.3	2.344	No	No	Nontoxic	0.231	No	No	Nontoxic
		2005	92	5.2	1.79	No	No	Nontoxic	2.068	No	No	Nontoxic
1.83	23	2000	89	11.4	2.3	No	No	Nontoxic				
		2004	90	4.08	2.282	No	No	Nontoxic	0.740	No	No	Nontoxic
		2005	88	2.7	3.09	yes	No	Nontoxic	0.229	No	No	Nontoxic
1.89	21	2000	93	6.7	0.23	No	No	Nontoxic				
		2004	88	6.45	3.307	Yes	No	Nontoxic	1.388	No	No	Nontoxic
		2005	95	5.5	-0.62	No	No	Nontoxic	2.558	No	No	Nontoxic
2.18	N1	2000	97	4.5	0.3	No	No	Nontoxic				
		2004	94	7.5	1.956	No	No	Nontoxic	0.658	No	No	Nontoxic
		2005	88	14.4	1.76	No	No	Nontoxic	1.099	No	No	Nontoxic
2.26	30	2000	97	2.7	0.96	No	No	Nontoxic				
		2004	92	12.6	0.837	No	No	Nontoxic	0.419	No	No	Nontoxic
		2005	82	5.2	0.98	No	No	Nontoxic	0.848	No	No	Nontoxic
2.58	24	2000	99	2.2	-0.24	No	No	Nontoxic				
		2004	93	7.5	1.173	No	No	Nontoxic	0.611	No	No	Nontoxic
		2005	95	4	0.68	No	No	Nontoxic	3.562	Yes	No	Nontoxic
2.71	15	2000	96	4.2	0.51	No	No	Nontoxic				
		2004	95	4.08	1.481	No	No	Nontoxic	1.047	No	No	Nontoxic
		2005	83	6.1	0.61	No	No	Nontoxic	0.729	No	No	Nontoxic
2.92	5	2000	96	6.5	0.6	No	No	Nontoxic				
		2004	90	3.54	1.649	No	No	Nontoxic	0.716	No	No	Nontoxic
		2005	98	2.6	-0.65	No	No	Nontoxic	5.156	Yes	No	Nontoxic
3.03	1	2000	97	4.5	0.34	No	No	Nontoxic				
		2004	91	10.3	1.939	Yes	No	Nontoxic	0.014	No	No	Nontoxic
		2005	88	8	2.17	No	No	Nontoxic	0.493	No	No	Nontoxic
3.14	28	2000	94	4.2	1.02	No	No	Nontoxic				
		2004	89	4.18	2.431	Yes	No	Nontoxic	0.942	No	No	Nontoxic
		2005	93	9.8	0.92	No	No	Nontoxic	3.187	Yes	No	Nontoxic

Distance (km) to Nearest Drill Centre	Station	Year	Amphipod Survival (%)	Sample Standard Deviation	Comparison to Laboratory Controls				Comparison to Reference Stations			
					Dunnett's t-stat	Statistically Significant	≥ 30% Reduction in Survival	Interpretation (Toxic / Nontoxic)	Dunnett's t-stat	Statistically Significant	≥ 20% Reduction in Survival	Interpretation (Toxic / Nontoxic)
3.18	25	2000	88	11.5	2.4	No	No	Nontoxic				
		2004	96	5.48	1.185	No	No	Nontoxic	1.333	No	No	Nontoxic
		2005	98	2.6		No	No	Nontoxic	4.329	Yes	No	Nontoxic
4.14	10	2000	95	5	1.01	No	No	Nontoxic				
		2004	89	6.29	3.014	Yes	No	Nontoxic	1.085	No	No	Nontoxic
		2005	82	11.3	-0.48	No	No	Nontoxic	0.829	No	No	Nontoxic
4.36	6	2000	97	2.7	0.3	No	No	Nontoxic				
		2004	94	4.18	0.977	No	No	Nontoxic	0.601	No	No	Nontoxic
		2005	94	4.9	0.89	No	No	Nontoxic	3.168	Yes	No	Nontoxic
4.45	18	2000	98	4.5	0	No	No	Nontoxic				
		2004	93	2.74	1.822	No	No	Nontoxic	0.254	No	No	Nontoxic
		2005	84	4.9	0.38	No	No	Nontoxic	0.425	No	No	Nontoxic
4.81	29	2000	93	10.3	1.28	No	No	Nontoxic				
		2004	89	5.48	4.056	Yes	No	Nontoxic	0.909	No	No	Nontoxic
		2005	94	4.9	0.69	No	No	Nontoxic	2.650	No	No	Nontoxic
4.88	2	2000	97	2.7	0.03	No	No	Nontoxic				
		2004	91	6.52	1.939	No	No	Nontoxic	0.280	No	No	Nontoxic
		2005	92	4.1	1.88	No	No	Nontoxic	1.982	No	No	Nontoxic
7.69	3	2000	94	5.5	1.35	No	No	Nontoxic				
		2004	75	14.1	5.451	Yes	No	Nontoxic	3.742	Yes	No	Nontoxic
		2005	94	4.9	0.9	No	No	Nontoxic	3.156	Yes	No	Nontoxic
7.89	22	2000	99	2.2	-0.34	No	No	Nontoxic				
		2004	93	5	1.458	No	No	Nontoxic	0.139	No	No	Nontoxic
		2005	98	2.6	-0.47	No	No	Nontoxic	4.329	Yes	No	Nontoxic
9.84	7	2000	95	5	0.91	No	No	Nontoxic				
		2004	95	4.08	0.721	No	No	Nontoxic	0.866	No	No	Nontoxic
		2005	82	8.2	0.95	No	No	Nontoxic	1.051	No	No	Nontoxic
10.26	26	2000	95	3.5	0.78	No	No	Nontoxic				
		2004	87	8.37	3.38	Yes	No	Nontoxic	1.255	No	No	Nontoxic
		2005	82	9.8	0.79	No	No	Nontoxic	0.767	No	No	Nontoxic
13.11	11	2000	96	4.2	0.51	No	No	Nontoxic				
		2004	99	2.24	1.776	No	No	Nontoxic	2.947	yes	No	Nontoxic
		2005	90	3.4	-0.94	No	No	Nontoxic	1.174	No	No	Nontoxic
20.03	27	2000	NS									
		2004	90	7.07	2.345	No	No	Nontoxic				
		2005	90	6.3	-1.2	No	No	Nontoxic				
22.62	12	2000	NS									
		2004	88	8.37	3.37	Yes	No	Nontoxic				
		2005	87	5.2	-0.23	No	No	Nontoxic				
22.95	4	2000	NS									
		2004	94	2.24	2.005	No	No	Nontoxic				
		2005	85	4.5	0.24	No	No	Nontoxic				
23.66	19	2000	NS									
		2004	99	2.24	0	No	No	Nontoxic				
		2005	83	6.1	0.61	No	No	Nontoxic				

Note: - NS = Not Sampled

Analysis of sediments kept with sediments tested for amphipod toxicity (refer to Section 5.2.2) indicate that holding time probably did not affect most sediment chemistry characteristics. More detailed results are provided in Appendix B-7. The most relevant finding was that ammonia levels decreased in sediments during storage. There was also evidence that ammonia levels in water overlying test sediment decreased during the 10 day toxicity tests (Appendix B-7).

Over all 44 stations sampled in 2005, amphipod survival decreased significantly with distances from the Northern and Central drill centres and was uncorrelated with sediment physical and chemical characteristics (Table 5-17). Stations 9 and N3 were not unusual (i.e., extreme) with respect to sediment physical and chemical characteristics (Table 5-18). The two stations were closer to drill centres than most other stations, but if there were drilling effects on the test amphipods they should also have occurred at other stations within 1 to 2 km of the drill centres.

**Table 5-17 Spearman Rank Correlations ( $r_s$ ) Between Amphipod Survival, Distances from the Drill Centres, and Sediment Physical and Chemical Characteristics (2005)**

Variable	Correlation ( $r_s$ ) with amphipod survival
Distance from:	
Northern drill centre	-0.356*
Central drill centre	-0.359*
Southern drill centre	-0.095
Nearest drill centre	-0.038
Barium	0.157
>C <sub>10</sub> -C <sub>21</sub> HCs	0.181
% fines	-0.143
TOC	0.049
Metals PC1	0.221
Ammonia	-0.085
Sulphide	-0.086
Sulphur	0.033
Redox	-0.197

Note: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)

**Table 5-18 Comparison of Distances from Drill Centres and Sediment Physical and Chemical Characteristics Between All Stations versus Stations 9 and N3 (2005)**

Variable	Minimum	Maximum	Median	Station 9	Station N3
Amphipod survival (%)	28	98	91	28	67.5
Distance (km) from:					
Northern drill centre	0.30	36.00	9.16	11.29	0.63
Central drill centre	0.33	29.66	4.51	4.58	7.68
Southern drill centre	0.59	30.00	4.66	1.61	10.52
Nearest drill centre	0.30	26.53	2.38	1.61	0.63
Barium (mg/kg)	93	810	170	240	220
>C <sub>10</sub> -C <sub>21</sub> HCs (mg/kg)	<0.3	260	1	3.1	11
% fines	0.77	2.67	1.20	1.43	0.86
TOC (g/kg)	0.6	1.1	0.9	0.9	0.9
Metals PC1	-2.78	3.84	0.28	0.47	-0.56
Ammonia (mg/kg)	2.3	49	7.3	8.6	7.8
Sulphide (mg/kg)	<0.2	1.0	<0.2	<0.2	<0.2
Sulphur (%)	0.018	0.048	0.025	0.021	0.024
Redox (mV)	154	334	248	198	280



5.4.3 Benthic Community Structure

A total of 15,745 invertebrates were collected from 44 stations in 2005, with mean abundances per station lower than in 2004 and 2000 (Table 5-19). The totals exclude nemerteans, nematodes, oligochaetes, ostracods and copepods. Over all three years, 102 “families” were collected. Some families were not taxonomic families, but represented individuals that could not be identified to family (e.g., Bivalvia unidentified), or higher taxonomic levels (e.g., phyla, class or order) that were not identified to lower levels. Raw data for benthic community structure are provided in Appendix B-4.

**Table 5-19 Taxonomic Composition of Benthic Invertebrate Community Samples (2000, 2004 and 2005)**

Phylum or subphylum	Class or order	No. families	2005 (EEM)		2004 (EEM)		2000 (baseline)	
			(n=44 stations)		(n=56 stations)		(n=44 stations)	
			No. organisms	% of total	No. organisms	% of total	No. organisms	% of total
Porifera		1	3	0.02	15	0.06	0	0.00
Cnidaria		6	24	0.15	160	0.63	13	0.04
Sipuncula		1	1	0.01	0	0.00	0	0.00
Annelida	Polychaeta	30	11,395	72.37	18,907	74.41	26,594	77.13
Mollusca	Total	33	2,939	18.67	4,368	17.19	5,930	17.20
	Bivalvia	18	2,870	18.23	4,290	16.88	5,857	16.99
	Gastropoda	15	69	0.44	78	0.31	73	0.21
Crustacea	Total	24	1,048	6.66	1,543	6.07	1,427	4.14
	Amphipoda	13	427	2.71	737	2.90	1,184	3.43
	Cirrepedia	1	20	0.13	2	0.01	13	0.04
	Cumacea	4	25	0.16	44	0.17	19	0.06
	Decapoda	1	0	0.00	0	0.00	1	0.00
	Isopoda	4	85	0.54	46	0.18	16	0.05
	Tanaidacea	1	491	3.12	714	2.81	194	0.56
Echinodermata		6	333	2.11	416	1.64	517	1.50
Urochordata	Ascidiacea	1	2	0.01	0	0.00	0	0.00
Total		102	15,745	100	25,409	100	34,481	100
Mean/station			358		454		750	

Note: - Stations represent two pooled samples, each approximately 0.1 m<sup>2</sup> in surface area

In all three years, polychaetes accounted for approximately 75% of the invertebrates collected, and bivalves accounted for 17 to 18% (Table 5-19). Therefore, these two higher-level (major) taxa accounted for 90% or more of the invertebrates collected. Amphipoda, Tanaidacea and Echinodermata were the only other major taxa accounting for more than 1% of total abundance in one or more years. Polychaetes and bivalves accounted for 48 of the 102 families collected. Fifteen (15) families of the relatively rare Gastropoda, and 13 families of Amphipoda, were collected.

In all three years, polychaetes in the family Spionidae (primarily *Prionospio steenstrupi*), were the most abundant (dominant) family (Table 5-20). Bivalves of the family Tellinidae (primarily *Macoma calcarea*, although juveniles can be difficult to identify to species) and polychaetes of the family Paraonidae (primarily *Aricidea catherinae*) were the second and third most abundant families. These three dominant taxa accounted for 60 to 70% of total abundance in each year. Therefore, invertebrate communities were not very rich or diverse even at the genus or species level.

Table 5-20 Dominant Benthic Invertebrate Families (2000, 2004 and 2005)

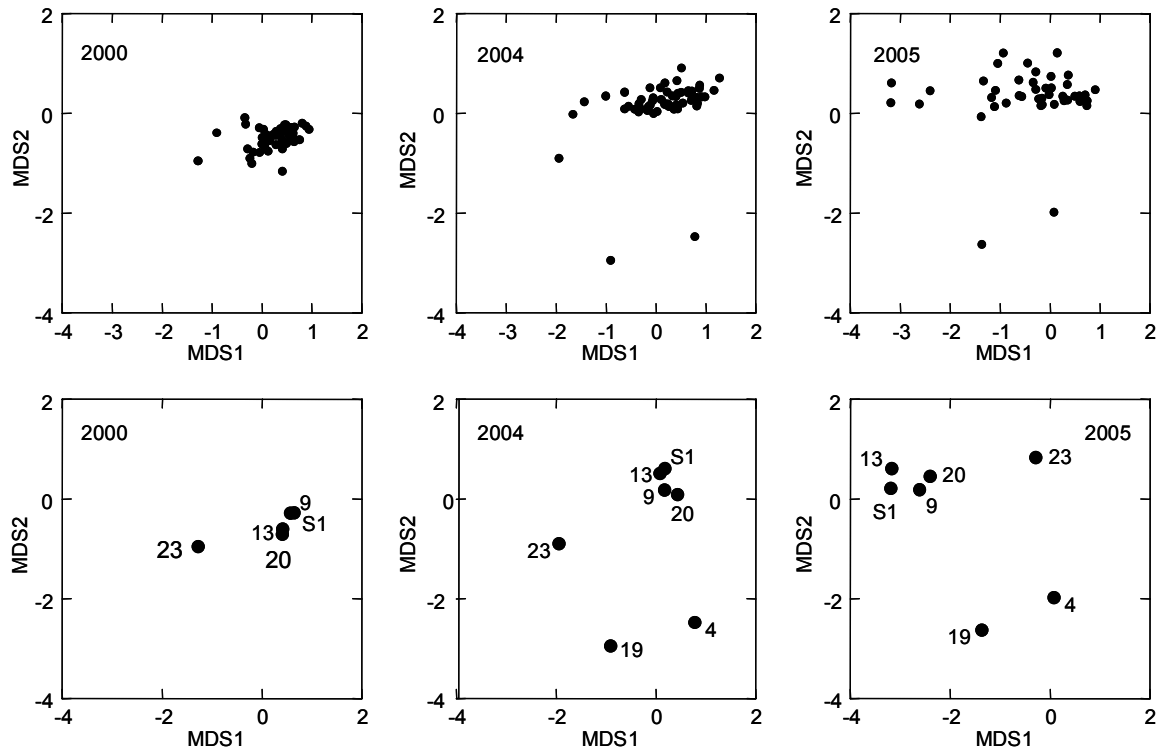
Major taxon	Family	2005				2004				2000			
		Abundance		Occurrence		Abundance		Occurrence		Abundance		Occurrence	
		No. organisms	% of total	No. stations	% of total	No. organisms	% of total	No. stations	% of total	No. organisms	% of total	No. stations	% of total
Polychaeta	Spionidae	5,736	36.4	44	100	9,462	37.2	56	100	12,812	37.2	46	100
Bivalvia	Tellinidae	2,456	15.6	44	100	3,784	14.9	56	100	4,616	13.4	46	100
Polychaeta	Paraonidae	2,307	14.7	41	93	5,004	19.7	56	100	5,020	14.6	46	100
Polychaeta	Orbiniidae	849	5.4	35	80	1,472	5.8	53	95	1,565	4.5	46	100
Tanaidacea		491	3.1	41	93	714	2.8	54	96	194	0.6	44	96
Polychaeta	Phyllodocidae	454	2.9	44	100	745	2.9	56	100	1,153	3.3	46	100
Polychaeta	Maldanidae	356	2.3	42	95	431	1.7	55	98	405	1.2	46	100
Polychaeta	Syllidae	353	2.2	33	75	524	2.1	52	93	312	0.9	44	96
Polychaeta	Cirratulidae	320	2.0	29	66	257	1.0	32	57	4,412	12.8	46	100
Echinodermata	Echinarachnidae	221	1.4	40	91	296	1.2	55	98	348	1.0	46	100
Polychaeta	Capitellidae	195	1.2	41	93	229	0.9	50	89	232	0.7	45	98
Amphipoda	Phoxocephalidae	150	1.0	25	57	182	0.7	43	77	269	0.8	43	93
Bivalvia	Hiatellidae	79	0.5	34	77	136	0.5	48	86	328	1.0	44	96
Amphipoda	Haustoriidae	54	0.3	20	45	227	0.9	50	89	641	1.9	46	100
Amphipoda	Dexaminidae	0	0.0	0	0	259	1.0	51	91	176	0.5	41	89
Bivalvia	Carditidae	0	0.0	0	0	0	0.0	0	0	443	1.3	42	91

Relative (%) abundances of most sub-dominants listed in Table 5-20 were similar among years. However, there were large differences in relative abundances among years for some families, especially Cirratulidae (primarily *Chaetozone setosa*; abundant in 2000 but not 2004 and 2005), Dexaminidae (*Guernea nordenskioldi*; collected in 2000 and 2004 but not 2005), and Carditidae (*Cyclocardia* spp., collected only in 2000). These differences do not appear to be taxonomic/taxonomist artifacts, since both taxonomists have easily identified these taxa when they occurred in the White Rose or Terra Nova monitoring programs. Instead, year-to-year climate (e.g., cumulative degree-days at time of sampling) and other natural differences among years may affect abundances of seasonal and short-lived taxa, despite a relatively fixed calendar sample time. Differences among years in the set of stations sampled will also affect the numbers of some taxa that were only abundant at one or a few stations.

5.4.3.1 Preliminary Analysis

Non-Metric Multidimensional Scaling

Figure 5-31 provides the two-dimensional NMDS plot based on relative abundances of invertebrate families for the 146 stations sampled in 2000, 2004 and 2005. The stress coefficient was 0.17, which represents a reasonable fit to the original pair-wise B-C similarity matrix (Clarke 1993). The top row in Figure 5-31 provides plots of all stations sampled in each year; the bottom row isolates stations that were unusual in one or more years.



**Figure 5-31 NMDS Plots Based on Relative (%) Abundances of Invertebrate Taxa (2000, 2004 and 2005)**

MDS1 scores were positively correlated with abundances of the dominant polychaete families, Spionidae and Paraonidae, and negatively correlated with abundances of the dominant bivalve family, Tellinidae. Abundances of some sub-dominant polychaete families (e.g., Maldanidae) were negatively correlated with MDS1 scores and positively correlated with Tellinidae abundances (i.e., not all polychaetes “behaved” similarly). Tellinidae accounted for most of the bivalves collected; variances of abundances of other bivalve taxa were mostly associated with variance along MDS2 and not MDS1.

MDS2 scores were strongly negatively correlated with relative abundances of Haustoriidae (Amphipoda), Carditidae (Bivalvia) and Cirratulidae (Polychaeta), three taxa that decreased substantially in abundance between 2000 and 2004/2005 (Table 5-20). MDS2 scores were positively correlated with abundances of Tanaidacea and a large number of rarer taxa that increased in abundance between 2000 and 2004/2005. Consequently, MDS2 scores for most stations increased from 2000 to 2004/2005 (Figure 5-31). In 2000, Cirratulidae were abundant at most stations and accounted for 13% of all invertebrates collected. In 2004 and 2005, Cirratulidae were among the dominants at reference stations 4 (Northeast) and 19 (Southwest), which were not sampled in 2000, but rare or absent at other stations. Stations 4 and 19 were outliers for MDS2, even relative to stations sampled in 2000 (Figure 5-31). The similarity in MDS2 scores for stations 4 and 19, and differences between these two stations and other stations, was surprising, since the two stations were the shallowest (station 19) and deepest (station 4) sampled, and separated by almost 60 km.

**Summary Statistics**

Table 5-21 provides summary statistics for invertebrate community summary measures, relative (%) abundances of major taxa, and absolute abundances of Amphipoda and Echinodermata, for the 44 stations sampled in 2005. Mean abundances varied 10-fold among stations with SD more than 50% of means. (i.e., CVs were greater than 50%) Other summary measures were less variable. Mean standing crop was 177 g wet/station, so mean wet weight per organism was approximately 0.5 g (ignoring the small contribution of excluded taxa to standing crop). Although more than 20 taxa were collected at most stations, diversity (number of dominant taxa per station) was much lower (3 to 7). Most stations were dominated by Spionidae, Paraonidae, Tellinidae plus one or a few of the sub-dominants in Table 5-21. Consequently, evenness values were low (mean and median less than 0.2 versus the maximum possible value of 1). As noted elsewhere, polychaetes and bivalves were the dominant major taxa. Absolute abundances of amphipods were highly variable with SD approximately equal to means. Absolute abundances of the rarer but larger echinoderms were less variable.

**Table 5-21 Summary Statistics for Benthic Invertebrate Community Variables (2005)**

Variable	Units	Min	Max	Median	Mean	SD	CV (%)
<i>Summary measures</i>							
Total abundance	No. organisms	75	1,197	329	358	201	56
Standing crop	g wet	62	308	167	177	67	38
Richness (S)	No. taxa	15	44	26	25	5	21
Diversity (D)	No. dominant taxa	3.3	6.7	4.7	4.7	0.8	17
Evenness (E)	D/S	0.12	0.34	0.18	0.19	0.05	27
MDS1		-3.19	0.90	-0.17	-0.42	1.00	
MDS2		-2.63	1.21	0.34	0.32	0.65	
<i>Major taxon abundances</i>							
% Polychaeta		30.0	86.7	69.9	67.4	13.8	20
% Bivalvia		3.2	56.0	19.7	22.5	12.2	54
% Amphipoda		0.0	6.6	2.2	2.4	1.7	69
% Echinodermata		0.0	12.2	1.9	2.8	2.7	95
% Tanaidacea		0.0	8.5	2.7	3.0	2.1	69
No. Amphipoda		0	55	8.5	9.7	9.8	101
No. Echinodermata		0	21	7.0	7.6	4.7	61

Notes: - All values were based on pooling two samples per station. Each sample was approximately 0.1 m<sup>2</sup> in surface area  
 - Richness, diversity, evenness and MDS scores were based on families

**5.4.3.2 Correlations Within and Among Groups of Variables (2005)**

**Correlations Among Invertebrate Community Variables**

Table 5-22 provides rank correlations ( $r_s$ ) among invertebrate community variables. Total abundance was strongly positively correlated with richness, which is typically the case, since more taxa will usually be collected when more organisms are collected. Abundance and diversity were uncorrelated, since the most abundant taxa were collected in most samples (Table 5-20). Richness and diversity were uncorrelated; instead, diversity was primarily a function of its other component, evenness. Despite the reasonably large number of taxa collected per station, diversity was low because abundances were unevenly distributed among these taxa. Evenness can be considered largely redundant, since it was calculated from diversity and richness (i.e.,  $E=D/S$ ). Results for evenness could usually be inferred from results for diversity and richness,

and these results were typically “no relationship/effects of interest” (i.e., evenness was an insensitive variable).

**Table 5-22 Spearman Rank Correlations ( $r_s$ ) Among Benthic Invertebrate Community Variables (2005)**

	Total abundance	Standing crop	Richness	Diversity	Evenness	MDS1	MDS2
Standing crop	0.196						
Richness	<b>0.777***</b>	0.110					
Diversity	-0.058	-0.095	0.034				
Evenness	<b>-0.674***</b>	-0.099	<b>-0.730***</b>	<b>0.572***</b>			
MDS1	<b>0.640***</b>	0.303*	0.465**	-0.184	<b>-0.524***</b>		
MDS2	-0.186	0.158	-0.091	-0.045	0.098	-0.010	
% Polychaeta	<b>0.734***</b>	0.329*	<b>0.516***</b>	-0.177	<b>-0.568***</b>	<b>0.846***</b>	-0.165
% Bivalvia	<b>-0.695***</b>	-0.358*	<b>-0.517***</b>	0.055	0.492**	<b>-0.841***</b>	0.106
% Amphipoda	0.341*	0.060	0.276	0.101	-0.178	0.446**	-0.174
% Echinodermata	-0.490**	0.179	-0.366*	0.016	0.355*	-0.363	0.223
% Tanaidacea	0.233	-0.062	0.313*	0.377*	0.030	0.103	0.229
No. Amphipoda	<b>0.688***</b>	0.182	<b>0.547***</b>	0.053	-0.423**	<b>0.615***</b>	-0.224
No. Echinodermata	0.202	0.391*	0.180	0.011	-0.159	0.146	0.021

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - Richness, diversity, evenness and MDS scores were based on families

Total abundance was positively correlated with relative abundances of polychaetes and negatively correlated with the relative abundance of bivalves (Table 5-22). These correlations were expected, since total abundance should be greater where the most abundant major taxon (polychaetes) was dominant. Total abundance was also positively correlated with MDS1 scores, which were effectively Spionidae + Paronidae:Tellinidae. Given the abundance of these three families (Table 5-20), MDS1 scores were also effectively Polychaeta:Bivalvia, as the correlations between MDS1 and relative abundances of polychaetes and bivalves in Table 5-22 indicate.

Standing crop was positively correlated with MDS1 scores and the abundance of polychaetes relative to bivalves (Table 5-22), although one might expect standing crop to be greater where heavier shelled organisms such as bivalves are more abundant. Standing crop was positively correlated with absolute abundances of the rare but large echinoderms.

MDS1 scores were positively correlated with richness and negatively correlated with evenness (Table 5-22). The number of taxa collected increased when more polychaetes were collected, but evenness decreased because of the dominance of a few taxa. MDS2 scores were uncorrelated with other variables, probably because the scores were primarily a reflection of the replacement of some sub-dominant and rare taxa by others over time.

Absolute and, to a lesser extent, relative abundance of amphipods was positively correlated with total and polychaete abundances (Table 5-22). Correlations between amphipod abundances and other variables were largely indirect effects of the amphipod-polychaete correlation rather than any direct effects or contribution of amphipods themselves. Both amphipod and polychaete abundances were negatively affected by

drilling (see below). In this report, absolute rather than relative abundance of amphipods was analyzed because the absolute abundances provided a better measure of effects on amphipods alone.

The relative abundance of echinoderms was basically the inverse of total abundance (note the negative correlation in Table 5-22). The absolute abundance of echinoderms was uncorrelated with other variables except standing crop. Consequently, variance of % Echinodermata was largely a function of variance of, and effects on, total abundance (the denominator of % Echinodermata).

**Correlations Between Invertebrate Community Variables and Sediment Physical and Chemical Characteristics**

Tables 5-23 to 5-25 provide rank correlations between invertebrate community variables and sediment physical and chemical characteristics for stations sampled in 2005. Selected relationships for all three sample years are analyzed in more detail in Section 5.4.4. Invertebrate community variables were largely uncorrelated with sediment particle size and TOC (Table 5-23). The only significant correlation was a positive correlation between diversity and gravel content, which may indicate that there was a greater diversity of habitat (i.e., interstitial spaces of different sizes) where gravel content was higher. Total abundance, MDS1 scores and amphipod abundances were negatively correlated, and evenness and MDS2 scores positively (but weakly) correlated, with concentrations of the two drilling mud tracers, barium and >C<sub>10</sub>-C<sub>21</sub> HCs (Table 5-24). The community variables were uncorrelated with Metals PC1 (i.e., concentrations of metals other than barium) (Table 5-24) and ammonia concentrations (Table 5-25). Correlations with sulphur, a potential tracer of drilling muds, were similar to those for barium and >C<sub>10</sub>-C<sub>21</sub> HCs (Table 5-25). Correlations with redox, which generally increased with increasing distance from drill centres and decreasing tracer concentrations, were of opposite sign to correlations with sulphur, barium and >C<sub>10</sub>-C<sub>21</sub> HCs. Collectively, these results indicate that the sediment characteristics most affected by drilling had the strongest relationships with invertebrate community variables.

**Table 5-23 Spearman Rank Correlations (*r<sub>s</sub>*) Between Benthic Invertebrate Community Variables and Sediment Particle Size and TOC (2005)**

Benthic invertebrate community variable	Sediment particle size and organic carbon content		
	% fines	% gravel	TOC
Total abundance	-0.150	0.084	-0.075
Standing crop	-0.123	-0.139	-0.166
Richness	-0.151	0.118	-0.080
Diversity	0.101	0.304*	0.060
Evenness	0.145	-0.029	0.139
MDS1	-0.216	-0.213	0.000
MDS2	-0.039	0.066	-0.288
No. Amphipoda	-0.051	-0.056	0.249

Notes: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)  
 - Richness, diversity, evenness and MDS scores were based on families

**Table 5-24 Spearman Rank Correlations ( $r_s$ ) Between Benthic Invertebrate Community Variables and Barium, >C<sub>10</sub>-C<sub>21</sub> HCs and Metals PC1 (2005)**

Benthic invertebrate community variable	Sediment chemistry variable		
	Barium	>C <sub>10</sub> -C <sub>21</sub> HCs	Metals PC1
Total abundance	-0.473**	<b>-0.534***</b>	0.024
Standing crop	-0.048	-0.075	-0.189
Richness	-0.316*	-0.313*	0.018
Diversity	0.113	-0.087	0.062
Evenness	0.350*	0.272	0.036
MDS1	<b>-0.579***</b>	<b>-0.515***</b>	-0.046
MDS2	0.305*	0.419**	-0.061
No. Amphipoda	<b>-0.573***</b>	<b>-0.729***</b>	0.002

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - Richness, diversity, evenness and MDS scores were based on families

**Table 5-25 Spearman Rank Correlations ( $r_s$ ) Between Benthic Invertebrate Community Variables and Ammonia, Sulphur and Redox (2005)**

Benthic invertebrate community variable	Sediment chemistry variable		
	Ammonia	Sulphur	Redox
Total abundance	0.088	<b>-0.528***</b>	0.413**
Standing crop	-0.178	-0.106	0.041
Richness	0.156	-0.457**	0.463**
Diversity	0.141	-0.007	-0.018
Evenness	0.020	0.339*	-0.360*
MDS1	-0.208	-0.340*	0.346*
MDS2	-0.216	0.026	-0.205
No. Amphipoda	0.125	-0.380*	<b>0.509***</b>

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - Richness, diversity, evenness and MDS scores were based on families

**Correlations Between Invertebrate Community Variables and Amphipod Survival**

Over all 44 stations, amphipod survival in toxicity tests was not significantly correlated with any of the eight benthic invertebrate community variables. However, at station 9 and, to a lesser extent, station N3 where amphipod survival in toxicity tests was lower than in other samples, total abundance, MDS1 scores and amphipod abundance were reduced (Table 5-26). This is evidence of agreement between laboratory and field responses, especially for amphipods, for the two stations, but there were several to many other stations where effects were observed in the field but not in the laboratory tests.

**5.4.3.3 Depth and Distance Effects (2005)**

Table 5-27 provides results of rank-rank regressions of invertebrate community variables on depth and distances from the drill centres for stations sampled in 2005. Overall multiple correlations ( $R$ ) for regression models with more than one  $X$  variable can range from 0 to 1. Partial correlations ( $r$ ) for each  $X$  variable can range from -1 to 1, and provide the correlation between the  $X$  variable and  $Y$  with the effects of other  $X$  variables held constant.

**Table 5-26 Comparison of Benthic Invertebrate Community Variable Values for All Stations versus Stations 9 and N3 (2005)**

Variable	Minimum	Maximum	Median	Station 9	Station N3
Amphipod survival (%)	28	98	91	28	67.5
Total abundance	75	1,197	329	104	134
Standing crop	62	308	167	148	165
Richness	15	44	26	15	21
Diversity	3.3	6.7	4.7	5.0	4.5
Evenness	0.12	0.34	0.18	0.34	0.22
MDS1	-3.19	0.90	-0.17	-2.61	-1.34
MDS2	-2.63	1.21	0.34	0.18	0.65
No. Amphipoda	0	55	8.5	0	4

Note: - Richness, diversity, evenness and MDS scores were based on families

**Table 5-27 Results of Rank-Rank Regressions of Benthic Invertebrate Community Variables on Depth and Distances from the Drill Centres (2005)**

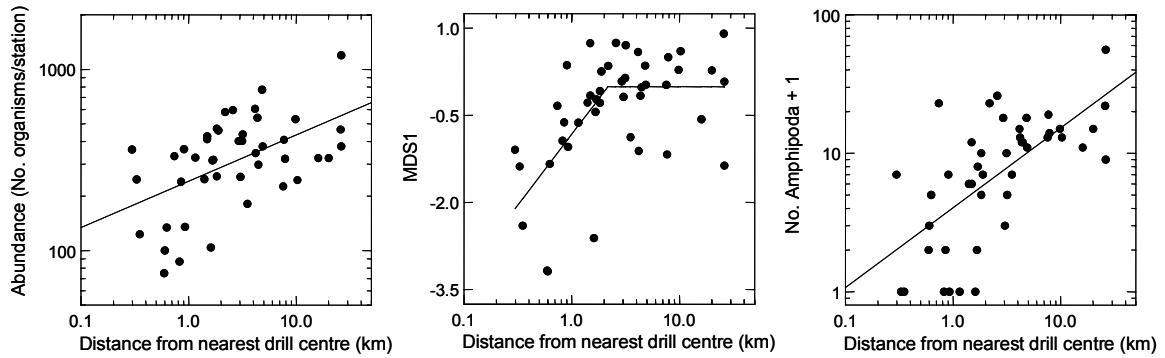
Y Variable	X=Depth & distances from each drill centre					X=Depth & distance from nearest drill centre (Min d)			X=Depth	X=Min d
	Overall R	Partial r				Overall R	Partial r		r <sub>s</sub>	r <sub>s</sub>
		Depth	N d	C d	S d		Depth	Min d		
Abundance	0.587**	0.415**	0.055	-0.315*	0.477**	0.500**	0.180	0.428**	0.286	0.474**
Standing crop	0.270	-0.081	-0.037	-0.062	0.166	0.122	-0.122	0.034	-0.117	-0.000
Richness	0.437	0.309*	-0.003	-0.250	0.340*	0.296	0.144	0.216	0.207	0.261
Diversity	0.413	0.409**	0.157	-0.250	0.207	0.351	0.305*	0.096	0.339*	0.183
Evenness	0.273	0.007	0.011	0.052	-0.181	0.180	0.076	-0.178	0.025	-0.164
MDS1	0.426	0.014	0.068	-0.144	0.333*	0.532**	-0.214	<b>0.531***</b>	-0.037	0.498**
MDS2	0.358	-0.165	-0.081	0.024	-0.182	0.333	-0.106	-0.281	-0.186	-0.318*
No. Amphipoda	<b>0.676***</b>	0.296	0.161	-0.141	0.486**	<b>0.667***</b>	0.071	<b>0.641***</b>	0.239	<b>0.665***</b>

Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - N, C and S d = distances from the Northern, Central and Southern drill centres  
 - Min d = distance from the nearest drill centre  
 - All Y and X variables were rank-transformed  
 - Richness, diversity, evenness and MDS scores were based on families

**Total Abundance, MDS1, Amphipod Abundance**

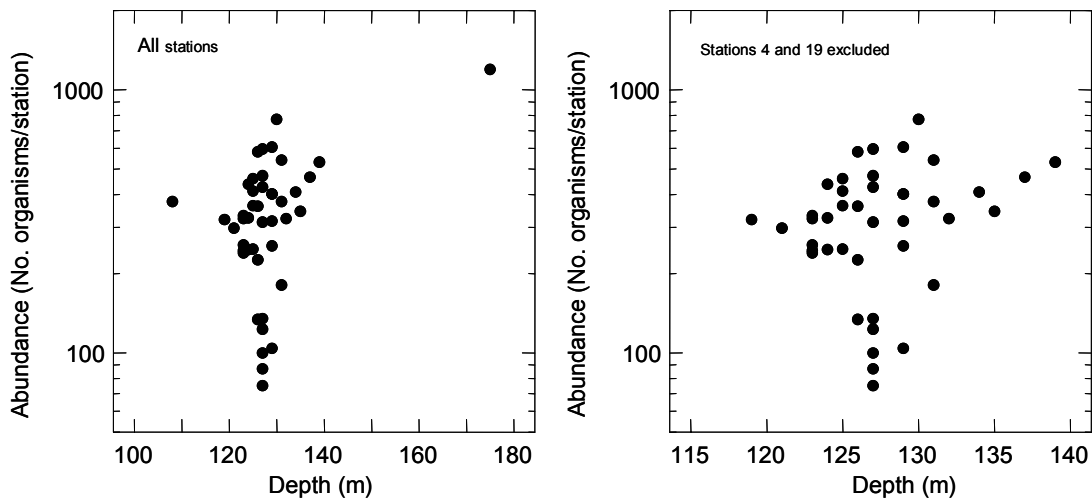
Total abundance, MDS1 scores (polychaete dominance) and amphipod abundances increased significantly with increasing distance from the nearest drill centre (Table 5-27; Figure 5-32). Distance from the nearest drill centre was used in plots and for several other purposes, but for these three variables, distance from the Southern drill centre alone may have been a more appropriate distance variable. Note that all partial r for Southern d were positive and significant for the variables, partial r for Northern d were weak and not significant, and partial r for Central d were negative (Table 5-27). There was a natural tendency for values of these variables, particularly total abundance, to decrease with distance from the Central drill centre (Section 5.4.3.4), which may have obscured any effects from drilling at this centre.





**Figure 5-32 Total Abundance, MDS1 and Amphipoda Abundance versus Distance from the Nearest Drill Centre (2005)**

Determining the most appropriate spatial regression model for total abundance is always problematic. Spatial distributions were complex, because they were a function of natural and project-related effects that differed among taxa. In the rank-rank regressions with depth and distances from all three drill centres in Table 5-27, but not in reduced models, depth effects were significant. Depth effects were also significant in a full parametric model, with or without a log-transformation for depth. However, any parametric model that includes depth is strongly influenced by extreme depths at station 4 (108 m) and station 19 (175 m). Figure 5-33 plots the relationship between total abundance and depth, with and without stations 4 and 19. With these two stations excluded, depth effects were not significant in parametric models.



**Figure 5-33 Total Abundance versus Depth (2005)**

If depth effects are not considered, the bivariate log-log regression of total abundance on distance from the nearest drill centre in Figure 5-32 is probably the best distance model for all 44 stations. Adding a threshold to the regression on distance from the nearest drill centre, or using distances from each drill centre, did not significantly reduce error variance (Table 5.28). The bivariate regression equation is:

$$\log_{10} \text{ Total Abundance} = 2.383 (\pm 0.043) + 0.256 (\pm 0.065) \times \log_{10} \text{ Min } d$$

In this and other equations, the “±” values are SE. Approximate 95% Confidence Intervals (CI) are regression intercepts or slopes ± 2 SE.

**Table 5-28 Results for Parametric Distance Models for Total Abundance, MDS1 and Amphipod Abundance (2005)**

Result/Estimate	Total abundance		MDS1	Amphipod abundance
	All stations	Stations 14 and 19 excluded	All stations	All stations
Regression on distance from nearest drill centre				
<i>r</i>	<b>0.520***</b>	0.468**	<b>0.486***</b>	<b>0.655***</b>
Full distance model				
Overall <i>R</i>	<b>0.580***</b>	0.543**	0.497**	<b>0.683***</b>
<i>p</i> for all drill centres vs nearest	0.149	0.141	0.756	0.259
Hockey-stick model				
Overall <i>R</i>	<b>0.556***</b>	<b>0.552***</b>	<b>0.617***</b>	<b>0.668***</b>
<i>p</i> for adding threshold ( $X_T$ )	0.136	0.034	0.004	0.268
antilog <i>a</i> (blade or background <i>Y</i> value)	392	376	-0.01 <sup>1</sup>	15.2 <sup>2</sup>
95% CI	316 to 486	301 to 469	-0.35 to 0.33 <sup>1</sup>	8.7 to 26.0 <sup>2</sup>
<i>b</i> (slope of shaft)	0.504 to 0.867	0.474 to 0.804	2.43 to 3.84	0.740 to 1.09
95% CI	0.141	0.144	1.02	0.394
antilog $X_T$ (threshold distance in km)	2.6	2.6	2.2	6.9
95% CI	1.1 to 6.3	1.0 to 6.7	1.1 to 4.2	2.4 to 19.5

- Notes:
- \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)
  - $X$  variables for the full distance model were distances from each drill centre
  - The  $X$  variable for the hockey-stick model was distance from the nearest drill centre
  - Both models were compared to bivariate regressions of  $Y$  on distance to the nearest drill centre
  - All  $X$  variables were log-transformed
  - Total abundance and amphipod abundance (+ 1) were log-transformed
  - <sup>1</sup>—Values are regression estimates without back-transformation (antilog), since MDS1 was not log-transformed
  - <sup>2</sup>—Values are antilogs minus 1

With stations 4 and 19 excluded, adding a threshold to the log-log relationship with distance to the nearest drill centre significantly reduced error variance, whereas using distance from each drill centre did not (Table 5-28). Without the highest abundance value at station 19 in Figure 5-32, a threshold relationship seems a reasonable fit. Estimates of zones of effects were the same ( $X_T = 2.6$  km), and 95% CI similar, regardless of whether stations 4 and 19 were included or excluded. Any decision about whether a bivariate versus hockey-stick relationship is more appropriate for the 2005 data depends largely on whether the high abundance at station 19 is considered a function of increased depth versus its distance from the drill centres.

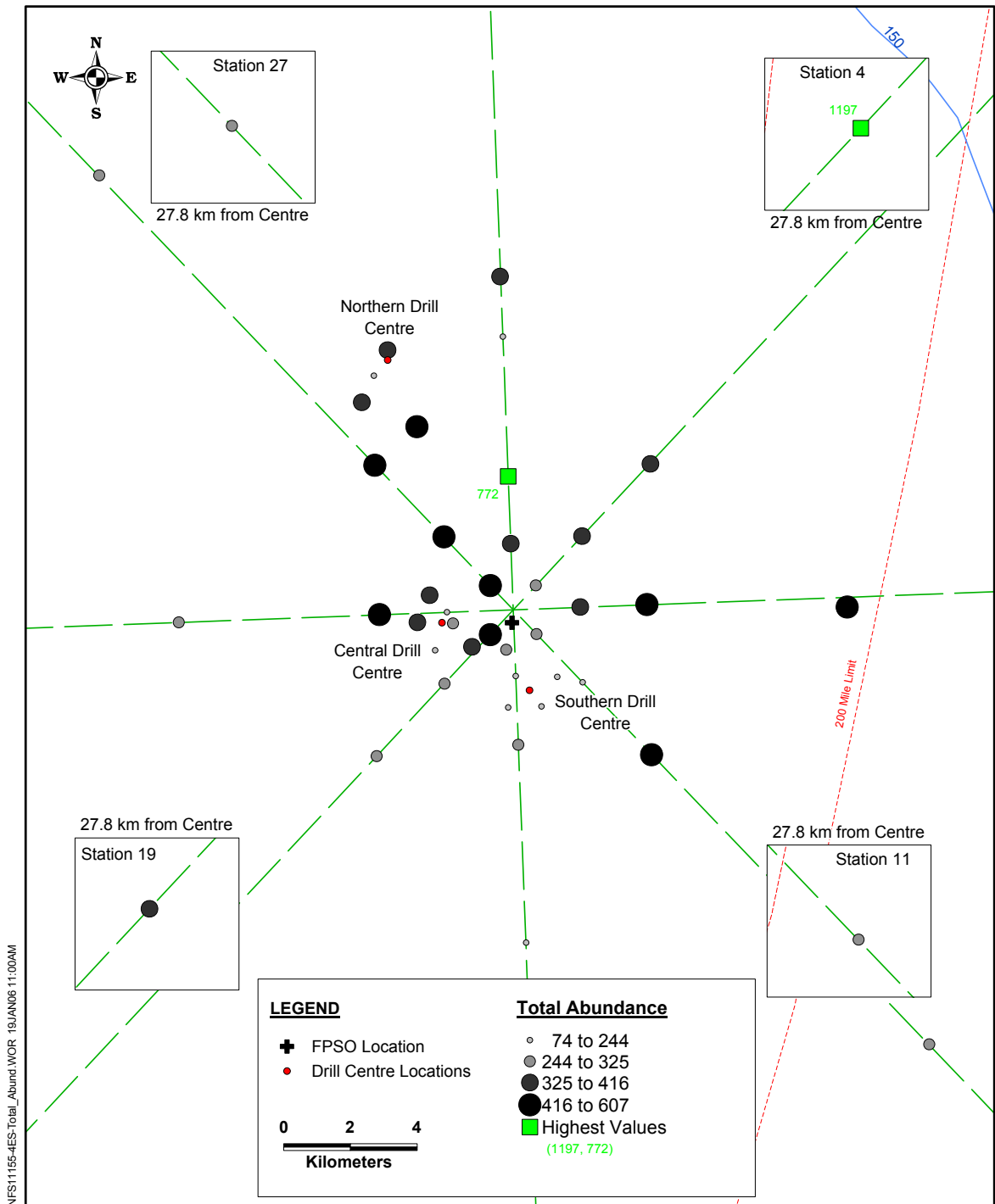
Figure 5-34 provides the spatial distribution of total abundance. Abundances were low in the immediate vicinity of the Central drill centres, but any effects did not appear to extend beyond 0.5 km. In contrast, abundances were substantially reduced out to at least 1 km southeast of the Southern drill centre. Abundances were not substantially reduced at stations between the Central and Southern drill centres. There was no apparent general increase in abundance from southwest to northeast along the depth gradient.

For MDS1, adding a threshold significantly reduced error variance relative to a bivariate linear regression on distance from the nearest drill centre, but using distances from each drill centre did not (Table 5-28). Table 5-28 provides the hockey-stick regression estimates which were used to plot the relationship in Figure 5-32. The estimated zone of effect was 2.2 km. Low MDS1 scores occurred near all three drill centres, with the lowest scores occurring at the two stations nearest the Southern drill centre (Figure 5-35). There were no apparent large-scale spatial patterns, consistent with the limited zone of effect estimate of 2.2 km.

For amphipod abundance, adding a threshold to a bivariate regression on distance from the nearest drill centre, or using distances from each drill centre, did not significantly reduce error variance (Table 5-28). The estimated threshold distance was 6.9 km, but the Upper 95% Confidence Limit (CL) was 19.5 km, near the upper end of the range of distances sampled. Therefore, a zone of effect, if defined, could include almost the entire sampling area. The regression equation for the bivariate model used in Figure 5-32 was:

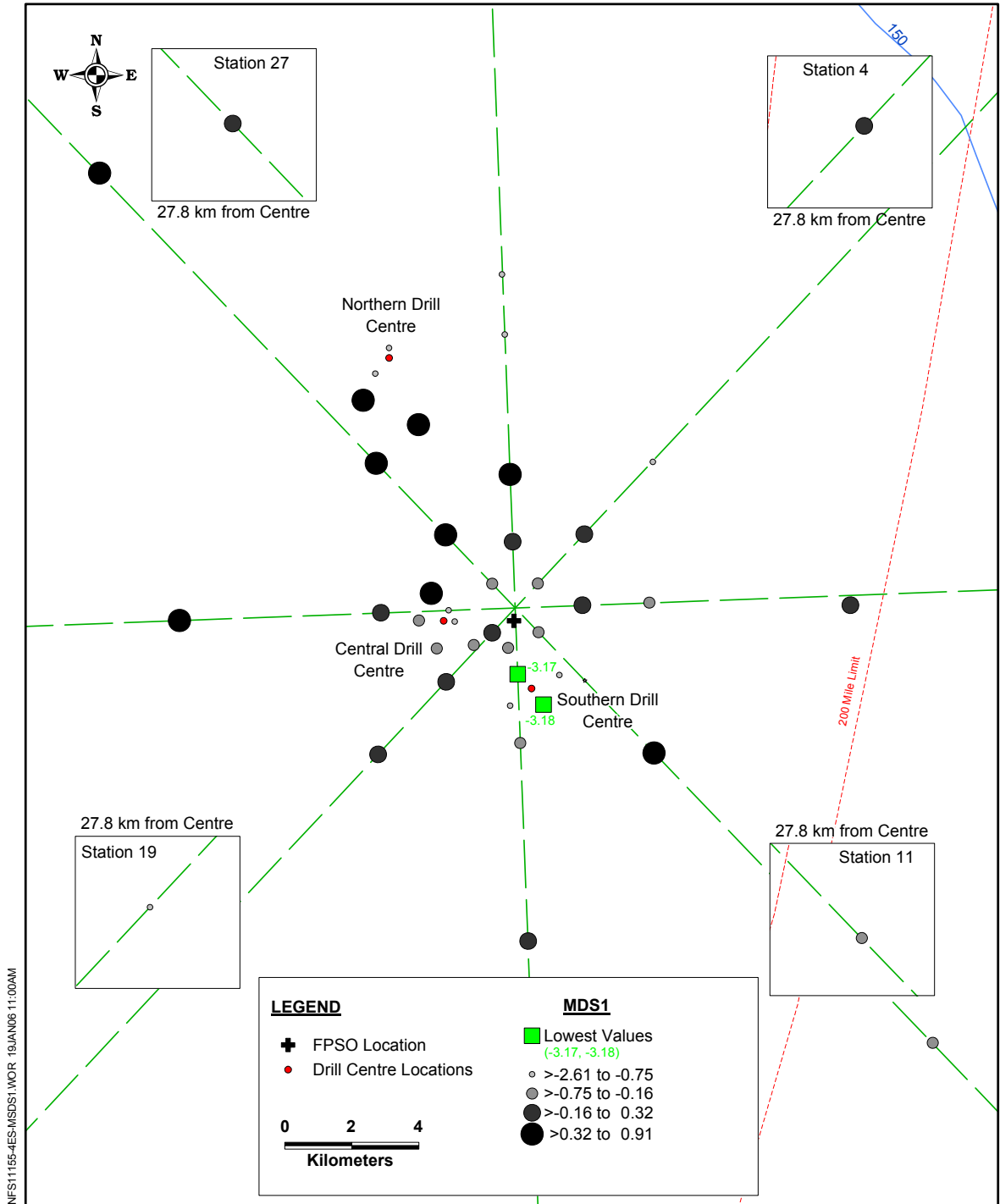
$$\log_{10} (\text{No. Amphipoda} + 1) = 0.606 (\pm 0.067) + 0.577 (\pm 0.103) \times \log_{10} \text{Min } d$$

Amphipod abundances were lowest (0 or near 0) at several stations near the Southern and Central drill centres (Figure 5-36). Reductions in abundance were generally greater to the southeast of the Central drill centre than to the northwest. Abundances were also lower near the Northern drill centre than at more remote stations.



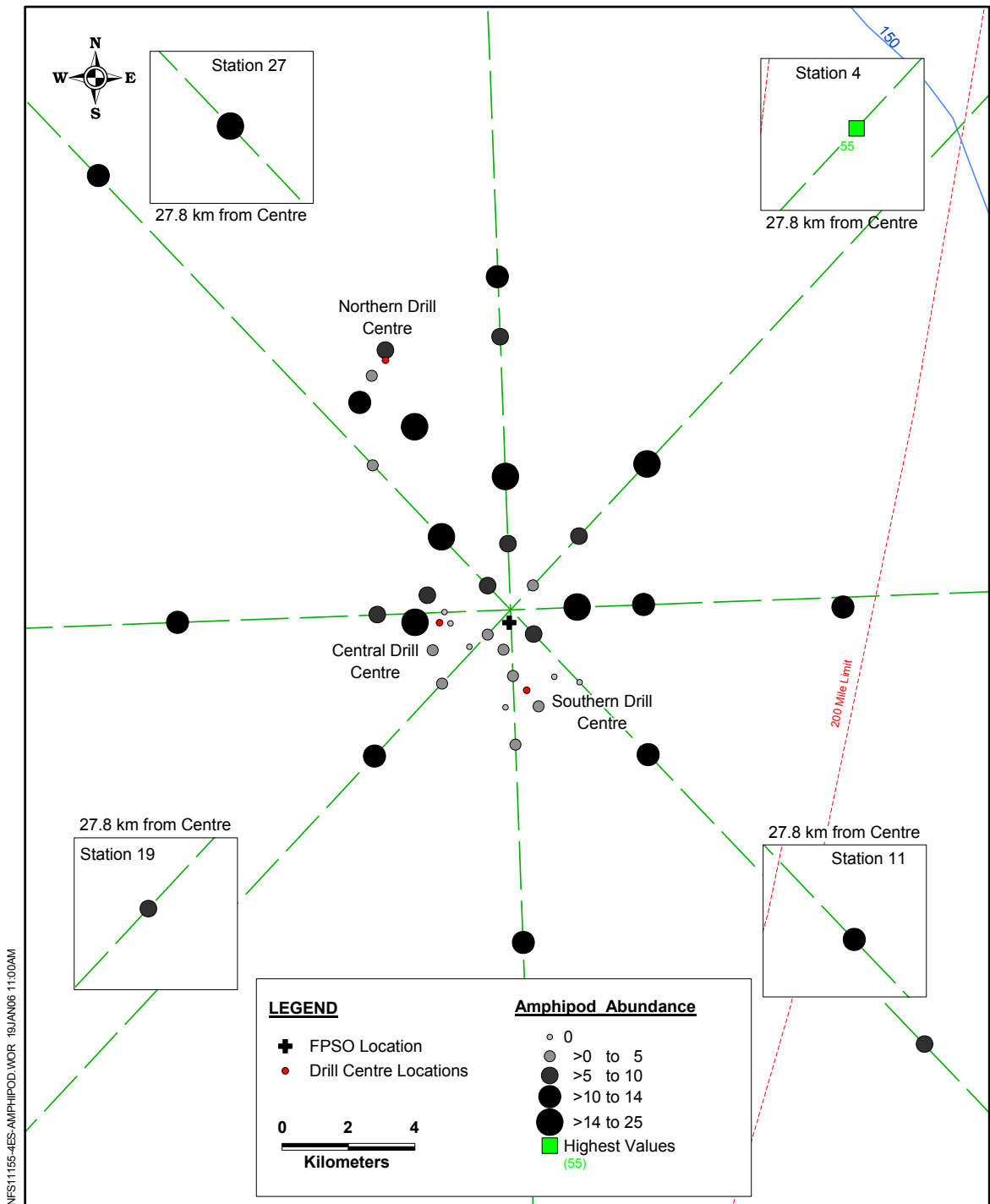
**Figure 5-34 Spatial Distribution of Abundance (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software



**Figure 5-35 Spatial Distribution of MDS1 (2005)**

Note: - Lowest values were identified as outliers by SPSS 14 software



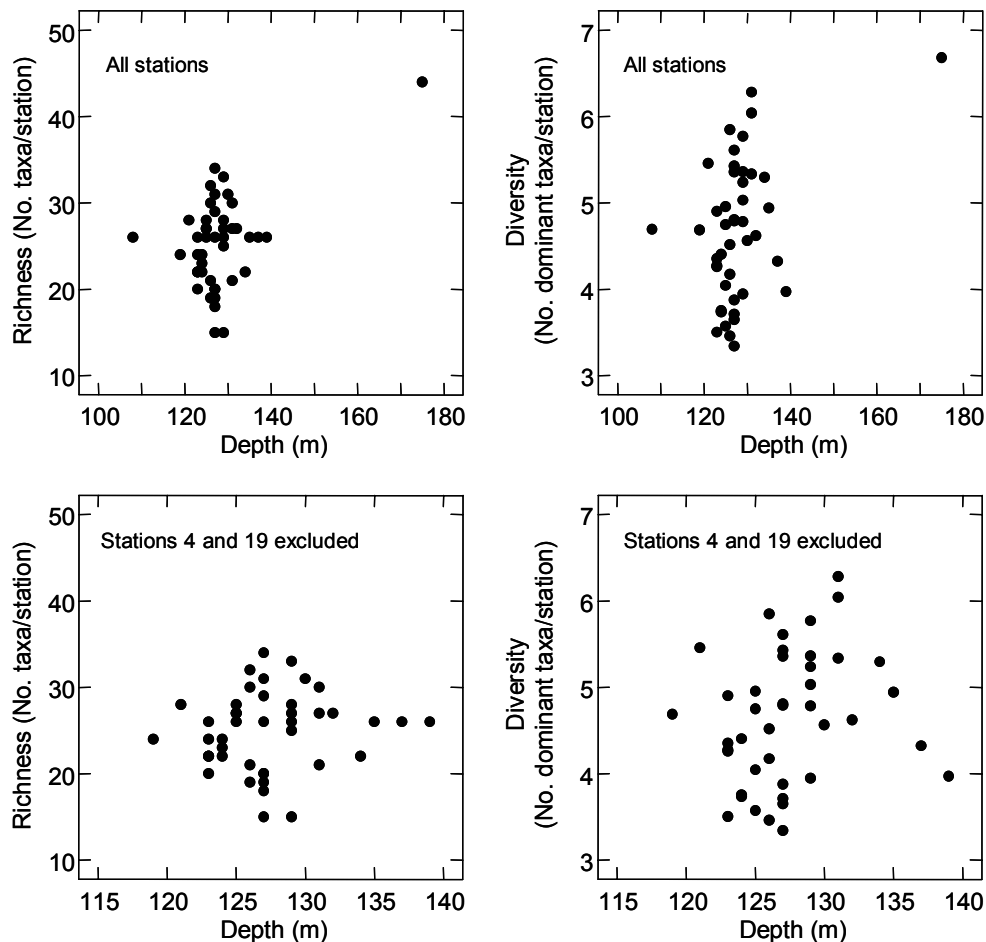
**Figure 5-36 Spatial Distribution of Amphipod Abundance (2005)**

Note: - Highest values were identified as outliers by SPSS 14 software

**Richness, Diversity and Evenness**

Depth and distance effects on richness were a mix of effects on total abundance and diversity. Richness was reduced at some stations near drill centres where abundance was reduced, but any distance effects on richness were weaker versions of effects on abundance (Table 5-27). Similarly, depth effects on richness were weaker versions of stronger depth effects on diversity, which was uncorrelated with abundance and largely unaffected by distance. There were no depth or distance effects on evenness.

Figure 5-37 plots relationships between richness and diversity versus depth. Depth effects for both variables were reduced by excluding stations 4 and 19 (extreme depth values). With these stations excluded, depth effects on diversity, but not richness, were still significant, with diversity increasing with increasing depth. The spatial distribution of diversity is plotted in Figure 5-38 as an example of a variable primarily affected by depth rather than distance. There was no apparent increase in diversity from southwest to northeast along the gradient of increasing depth; instead, there was a relatively wide variance of values near drill centres.



**Figure 5-37 Richness and Diversity versus Depth (2005)**

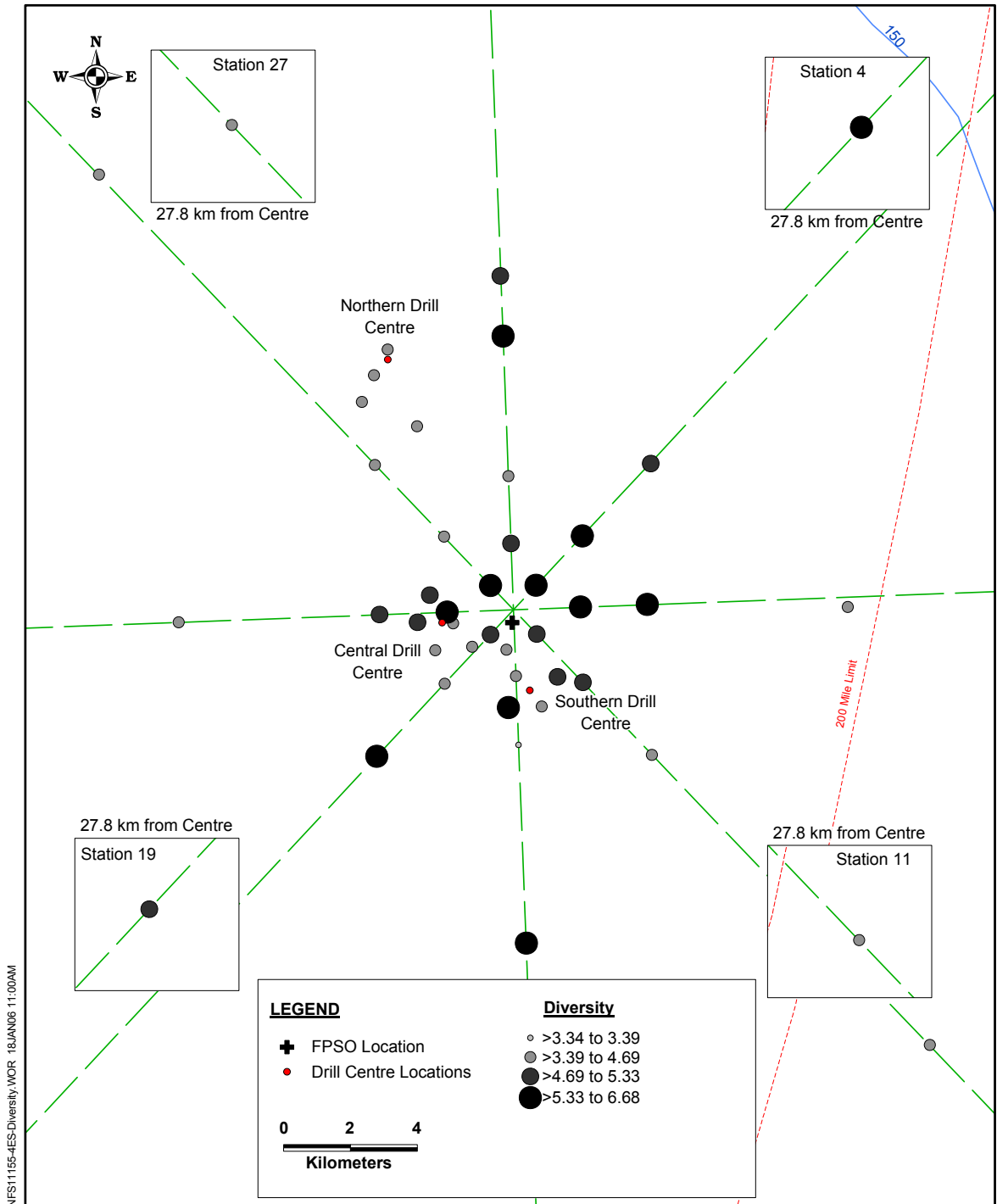


Figure 5-38 Spatial Distribution of Diversity (2005)



**Standing Crop and MDS2**

Standing crop and MDS2 are considered together as “left-over” variables (evenness could be added), with no apparent distance or depth effects on either variable (Table 5-27). Standing crop is mostly a function of the number of large but rare organisms collected, which probably varies randomly with respect to distance or depth.

Analyses of MDS2 scores are driven by the low scores at reference stations 4 and 19, and the variation in scores at other stations over time rather than space (Figure 5-31). In 2005, there was a significant negative correlation between MDS2 scores and distance from the nearest drill centre (Table 5-27). However, this correlation and any other distance effects were not significant with stations 4 and 19 (low MDS2 scores; high distance values) excluded. Depth effects will never be significant for MDS2 since stations 4 and 19 represent the highest and lowest depth values.

**5.4.3.4 Comparison Among Years (2000, 2004, 2005)**

Table 5-29 provides results of RM regression models comparing benthic invertebrate community variables among Years for the 37 stations sampled in all three sample years (2000, 2004 and 2005). Details of the analysis are provided in Appendix B-5; general guidelines for interpretation are provided in Section 5.4.1.3).

**Total Abundance, MDS1 and Amphipod Abundance**

Over all three sample years, total abundance increased with increasing depth (Among Stations Depth term in Table 5-29; Figure 5-39, top row). This relationship, presumably natural, complicated some other analyses of distance/project effects (see below, and Sections 5.4.3.3 and 5.4.4).

**Table 5-29 Results of RM Regression Analysis Comparing Benthic Invertebrate Community Variables Among 2000, 2004 and 2005**

Term	df	F value for Y variable							
		Abundance	Standing crop	Richness	Diversity	Even-ness	MDS1	MDS2	No. Amphipoda
<i>Among Stations</i>									
Depth	1,32	6.77*	0.57	10.08**	<b>15.78***</b>	0.23	10.55**	0.84	0.86
N d	1,32	1.24	0.03	3.06	1.19	0.32	0.28	0.05	0.18
C d	1,32	4.88*	0.11	6.68*	7.84**	0.08	0.24	3.34	1.69
S d	1,32	7.54**	2.34	2.15	1.18	0.73	4.39*	2.99	<b>15.75***</b>
Error 1 <sup>1</sup>	32,64	1.25	1.64*	1.06	0.65	0.56	1.46	0.96	1.49
<i>Within Stations</i>									
<i>Overall</i>									
Year	2,64	0.97	0.19	0.03	0.78	0.32	2.03	1.31	0.49
Year × Depth	2,64	1.01	0.25	0.16	0.55	0.24	1.80	0.56	0.03
Year × N d	2,64	0.01	1.02	1.07	4.45*	0.74	0.47	0.18	2.29
Year × C d	2,64	0.56	2.35	0.06	0.34	0.11	0.66	1.07	3.13
Year × S d	2,64	<b>16.28***</b>	0.02	2.81	0.61	1.80	<b>13.44***</b>	0.44	<b>16.13***</b>
<i>2000 versus 2004-05</i>									
Year	1,32	0.23	0.21	0.01	0.62	0.31	2.82	2.71	1.25
Year × Depth	1,32	1.10	0.08	0.18	0.24	0.27	3.80	1.13	0.07
Year × N d	1,32	0.02	0.78	2.07	5.48*	1.13	0.10	0.34	6.04*
Year × C d	1,32	0.21	0.57	0.01	0.14	0.18	1.82	0.07	0.27
Year × S d	1,32	<b>24.68***</b>	0.00	3.05	0.16	2.29	<b>18.69***</b>	0.50	<b>41.77***</b>
<i>2004 versus 2005</i>									
Year	1,32	1.55	0.17	0.05	1.14	0.32	1.67	0.00	0.03

Term	df	F value for Y variable							
		Abundance	Standing crop	Richness	Diversity	Even-ness	MDS1	MDS2	No. Amphipoda
Year × Depth	1,32	0.95	0.37	0.14	1.27	0.20	0.88	0.03	0.01
Year × N d	1,32	0.00	1.18	0.23	2.03	0.21	0.64	0.03	0.01
Year × C d	1,32	0.84	3.45	0.09	0.79	0.01	0.13	2.01	4.86*
Year × S d	1,32	9.75**	0.04	2.60	1.68	1.11	11.01**	0.38	0.52

Notes:

- Appendix B-5 explains terms and tests in the RM regression model
- df = degrees of freedom for the numerator (effect) and denominator (error) for *F*
- *d* = distances from various drill centres
- \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)
- *n* = 37 stations sampled in all three years
- Distances and all Y variables except Metals PC1 were log-transformed
- Richness, diversity, evenness and MDS scores were based on families
- <sup>1</sup>Error 1=carry-over effects or persistent differences among stations unrelated to depth or distance

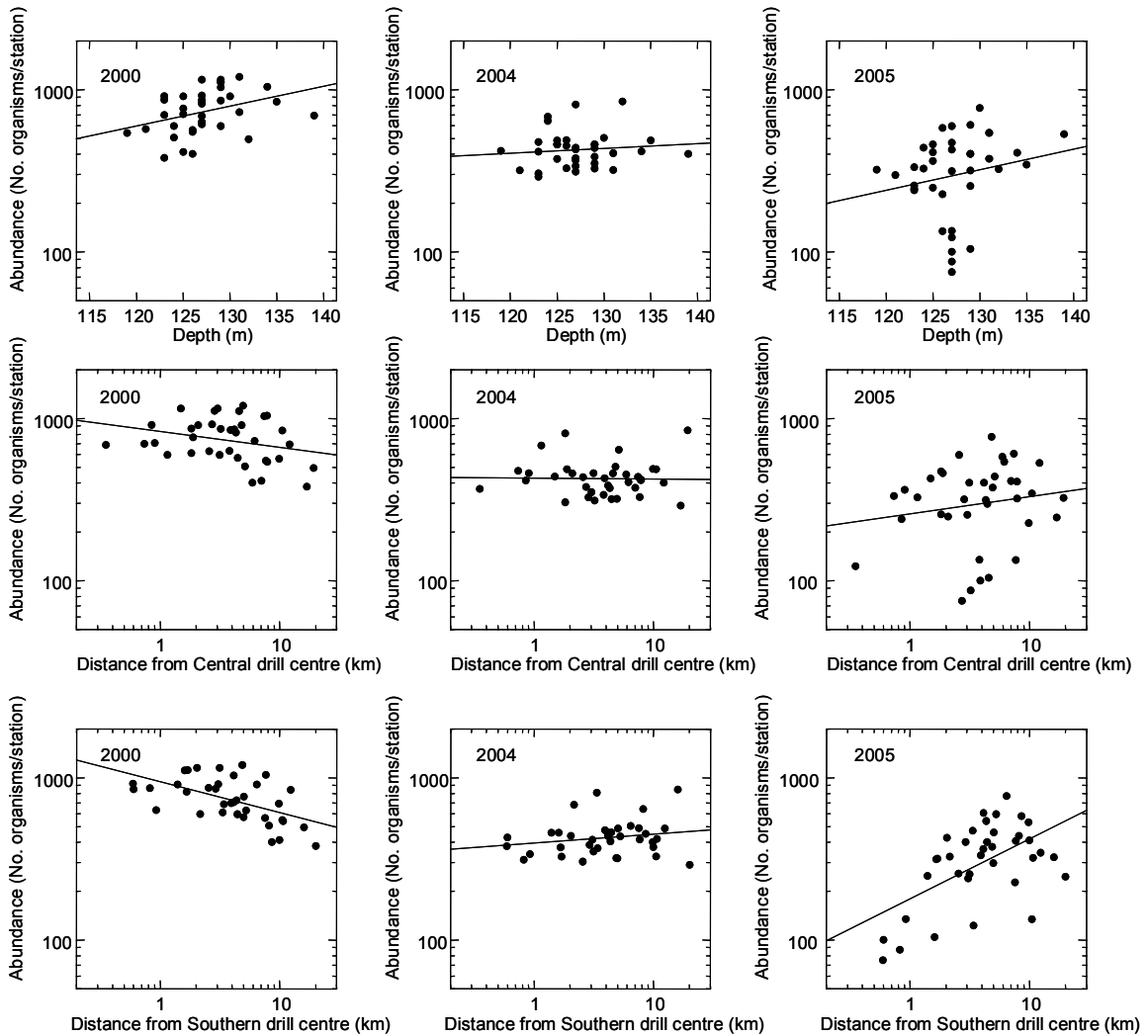
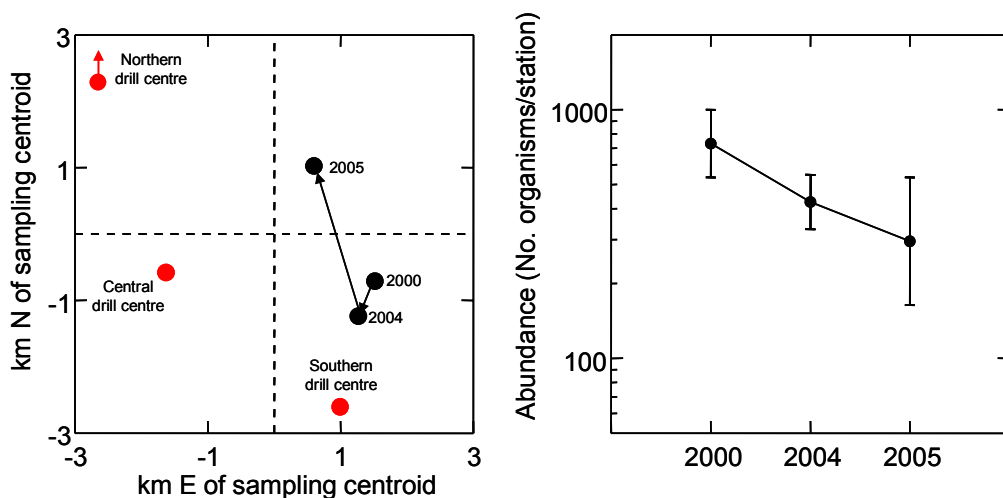


Figure 5-39 Total Abundance versus Depth and Distances from the Central and Southern Drill Centres (2000, 2004, 2005)

The relationship between total abundance and distance from the Southern drill centre progressively reversed over time (Figure 5-39). The differences in distance relationships between 2000 versus 2004 and 2005, and between 2004 versus 2005, were significant (Within Stations Year  $\times$  S  $d$  terms in Table 5-29). These results indicate that effects on abundance occurred after drilling began, and increased in strength in 2005 as drilling continued.

There was no evidence of effects from the Northern drill centre on total abundance, nor was there evidence of any natural gradient with distance from this drill centre. No Northern distance terms in the RM regression were significant (Table 5-29). The Among Stations test of distance from the Central drill centre was significant, but none of the Within Stations tests was significant. These results indicate that there was a natural gradient that has not changed over time. In 2000, abundance decreased with distance from the Central drill centre (Figure 5-39). This relationship appeared to weaken or even reverse over time, but this was largely because of effects from the nearby Southern drill centre (note the low abundances at intermediate distances). For these 37 stations, partial correlations for distance from the Central drill centre, with the effects of the other X variables removed, were -0.264, -0.209 and -0.267 for 2000, 2004 and 2005, based on the transformations used.

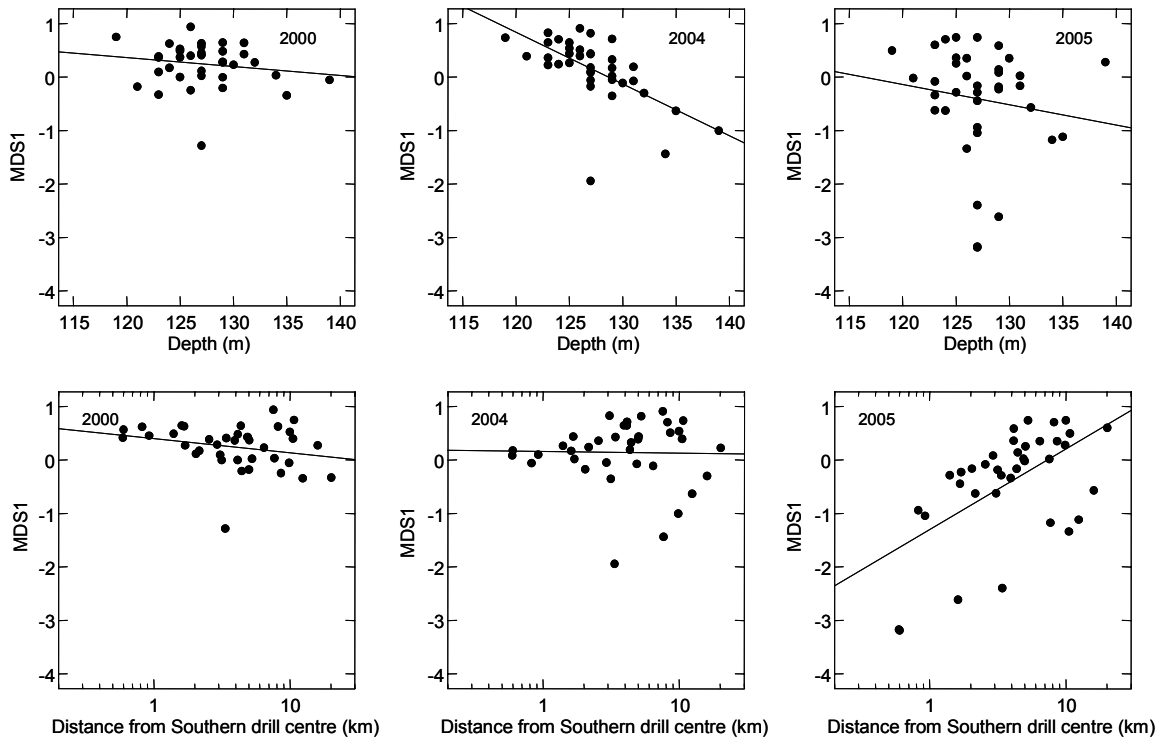
Sampling centroids for total abundance in 2000 and 2004 were located southeast of the sampling centroid (Figure 5-40). In 2005, the abundance centroid moved north and somewhat west (i.e., away from the Southern drill centre). The abundance centroids were all east of the sampling centroid because of depth effects (i.e., abundance increased with increasing depth from southwest to northeast). Mean abundance decreased over time (right plot in Figure 5-40). The decrease was partly natural, as abundance decreased somewhat at more remote stations between 2000 versus 2004 and 2005 (Figure 5-39), but was also attributable to some relatively large reductions in abundance near the Southern drill centre in 2004 and 2005.



**Figure 5-40 Total Abundance Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

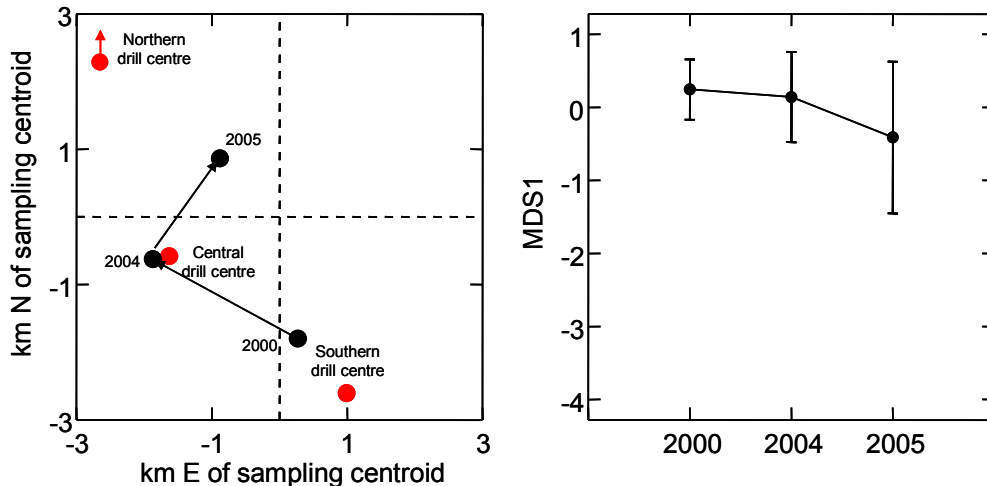
Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

Results for MDS1 were similar to those for total abundance, but with stronger depth effects and no natural relationship with distance from the Central drill centre (Table 5-29). MDS1 scores (i.e., polychaete dominance) decreased with increasing depth (Figure 5-41). Relationships with distance from the Southern drill centre reversed from weak decreases with distance in 2000 to relatively strong increases in 2005, with most of the change occurring between 2004 and 2005 rather than between 2000 and 2004. RM analysis of a similar community measure and the same 37 stations did not reveal any significant changes in the effects of distance from the Southern drill centre between 2000 and 2004 (Husky Energy 2005).



**Figure 5-41 MDS1 versus Depth and Distance from the Southern Drill Centre (2000, 2004, 2005)**

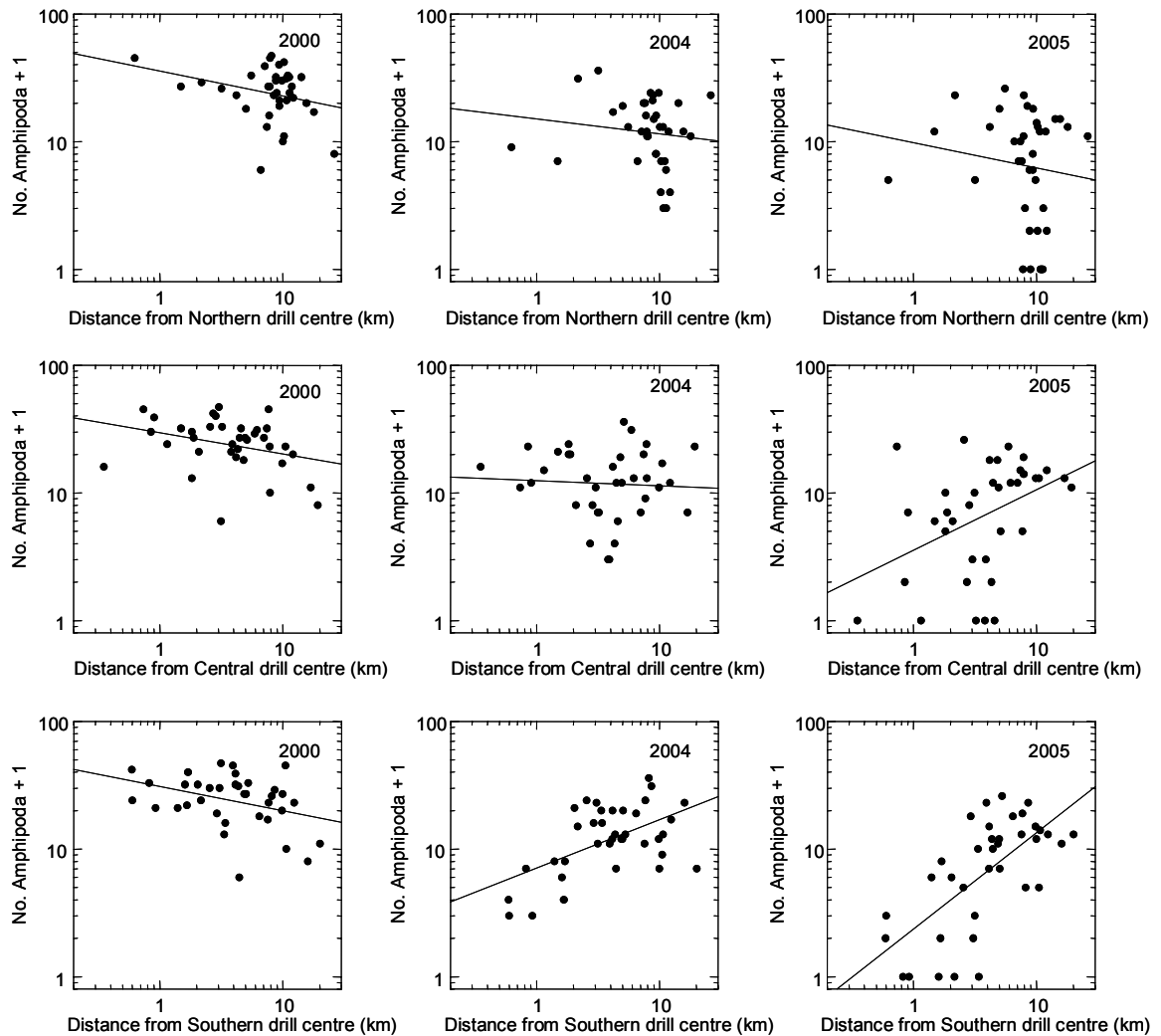
MDS1 centroids have progressively moved away from the Southern drill centre, possible evidence that there may have been some minor effects in 2004 (Figure 5-42). The centroids were generally located to the south and/or west of the sampling centroid (i.e., towards shallower depths). Average MDS1 scores have decreased over time, especially between 2004 and 2005. Some of this decrease may be natural, but some or even most of the decrease is attributable to effects of the Southern drill centre. For example, MDS1 scores at stations S1, 9 and 13 near the Southern drill centre were intermediate in 2000 and 2004, but were substantially reduced in 2005 to values lower than any observed in 2000 or 2004 (Figure 5-31).



**Figure 5-42 MDS1 Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

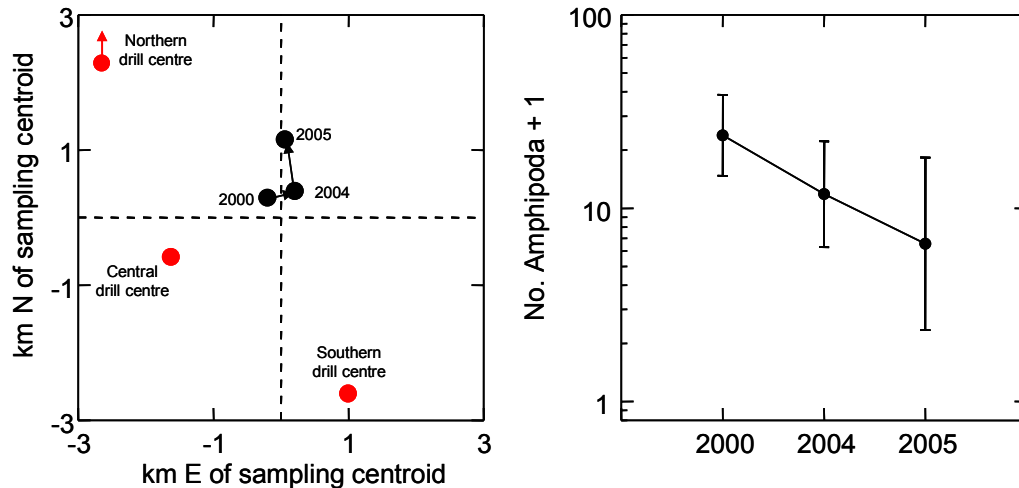
Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

RM analysis of amphipod abundances provided strong evidence of effects from all three drill centres (Table 5-29; Figure 5-43). In 2000, amphipod abundance decreased with distance from all three drill centres. Relationships between amphipod abundance and distances from the Northern and Southern drill centres reversed between 2000 and 2004, after drilling began at these two centres. Relationships between amphipod abundance and distance from the Central drill centre reversed between 2004 and 2005, after drilling began at this centre. All of these changes were significant (Table 5-29). These results indicated that amphipods were sensitive organisms, responding rapidly to drilling. Changes in distance gradients were also easier to detect (and visually apparent in Figure 5-43) than for other community variables, because depth effects were not significant, and natural distance gradients (decreases in amphipod abundance with distance) were opposite of those expected from drilling (increases with distance).



**Figure 5-43 Amphipod Abundance versus Distance from the Three Drill Centres (2000, 2004, 2005)**

The 2004 centroid for amphipod abundance did not move far from the 2000 centroid because abundance decreased with distance from both the Northern and Southern drill centres (i.e., the centroid remained mid-way between the two drill centres; Figure 5-44). In 2005, the centroid moved away from the Central and Southern drill centres. Amphipod abundances have progressively decreased over time (Figure 5-44). Some of the reduction may be natural, but most is attributable to drilling effects. Amphipod abundances at remote stations have not noticeably decreased, and 2005 was the first year in which abundances of 0 were observed near drill centres (Figure 5-43).



**Figure 5-44 Amphipod Abundance Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

Table 5-30 provides results and estimates for regressions of total abundance, MDS1 and amphipod abundance on distance from the nearest active drill centre (Southern and Northern) for all 56 stations sampled in 2004, with and without a threshold added. Direct and quantitative comparisons were not made to results for all 44 stations sampled in 2005 (Table 5-28) because distance relationships differed qualitatively between the two years. For total abundance and MDS1, regressions for 2004 should probably include depth, even with stations 4 and 19 excluded. Adding a threshold for regressions of total abundance on distance for 2004 did not significantly reduce error variance for total abundance, although the estimated threshold distance threshold (2.4 km) was similar to threshold distances estimated for 2005 (2.6 km). Adding a threshold reduced the error variance of distance models for MDS1, but any distance relationship was weak. Estimated distance thresholds were 0.6 km, and a function of reduced scores at a few stations near the drill centres. Again, that is evidence that limited and localized effects on MDS1 may have occurred in 2004, with these effects becoming more extensive in 2005. There was a significant threshold distance (2.8 km) for effects on amphipods in 2004, which was much lower than the estimate for 2005 (6.9 km with wide CI, and a bivariate regression is probably a better model than a hockey-stick regression). Therefore, the spatial extent of effects on amphipods increased in 2005 after drilling began at the Central drill centre.

**Table 5-30 Results for Parametric Regressions of Total Abundance, MDS1 and Amphipod Abundance on Distance from the Nearest Active Drill Centre (Northern and Southern) (2004)**

Result/Estimate	Total abundance		MDS1		Amphipod abundance
	All stations	Stations 4 and 19 excluded	All stations	Stations 4 and 19 excluded	All stations
Regression on distance from nearest active drill centre (Northern and Southern)					
<i>r</i>	0.322*	0.380*	0.172	0.221	<b>0.517***</b>
Hockey-stick model					
Overall <i>R</i>	0.340*	0.401*	0.324	0.338*	0.618
<i>p</i> for adding threshold	0.395	0.304	0.039	0.059	0.003
antilog $X_T$ (threshold distance in km)	12.7	2.4	0.6	0.6	2.8
95% CI	0.9 to 186	0.8 to 7.2	0.3 to 1.3	0.4 to 1.0	1.5 to 5.3

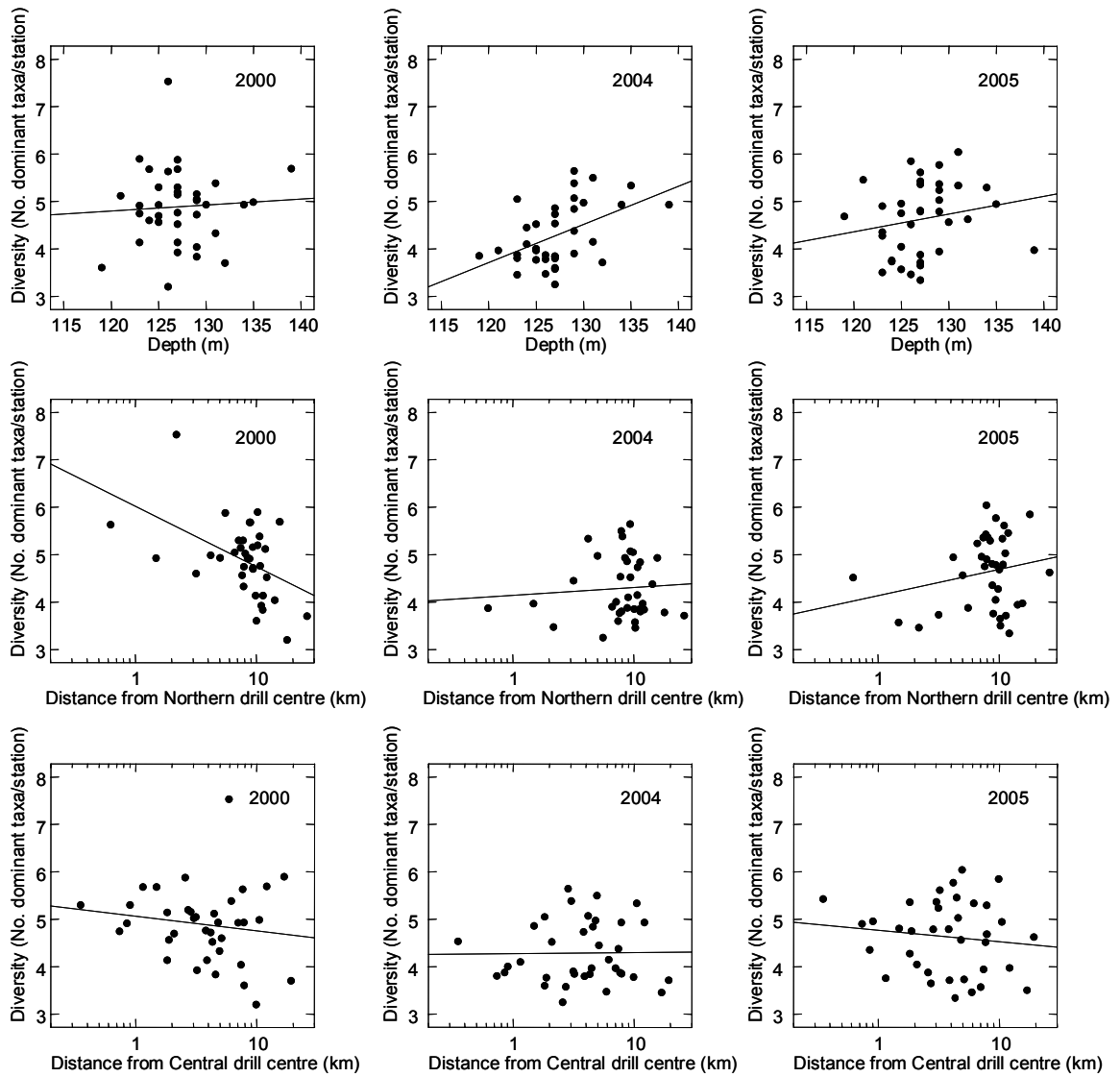
Notes: - \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - Distance, total abundance and amphipod abundance (+ 1) were log-transformed  
 -  $n = 56$  stations

**Richness, Diversity and Evenness**

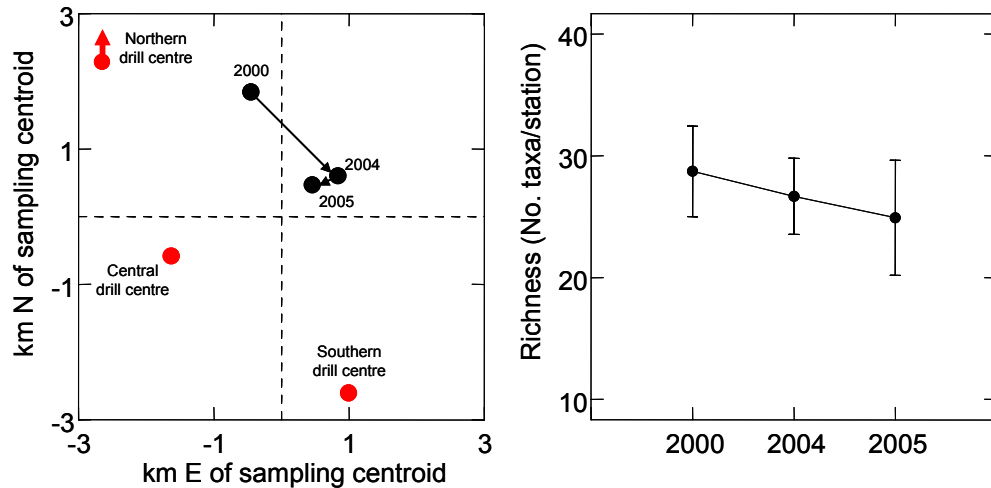
Richness and diversity increased significantly with increasing depth, and decreased with distance from the Central drill centre, in all three sample years (Among Stations terms in Table 5-29; Figure 5-45). Diversity only is plotted in Figure 5-45; relationships for richness were similar but weaker. The relationships between diversity and distance from the Northern drill centre also changed significantly between 2000 (no relationship) versus 2004 and 2005 (increase with distance) (Within Stations Year  $\times$  Northern *d* terms in Table 5-29). This could be evidence of adverse effects on diversity from drilling at the Northern drill centre; but if so, diversity would be the *only* variable affected by the Northern but not the Southern (or Central) drill centre. None of the terms or tests in the RM regression analysis in Table 5-29 was significant for evenness.

Changes in centroid locations and overall means over time were similar for richness and diversity (Figures 5-46 and 5-47). Centroids for both variables were to the north and/or east of the sampling centroid because of depth effects. Centroids have moved closer to the Southern drill centre over time, with these changes more apparent for diversity. Richness decreased slightly over time; diversity decreased in 2004 then returned to baseline (2000) levels in 2005. These overall changes over time were small and not significant (Within Stations Year terms in Table 5-29). Spatial changes in centroids for evenness were similar to those for diversity, and evenness values increased slightly but not significantly over time (Figure 5-48).



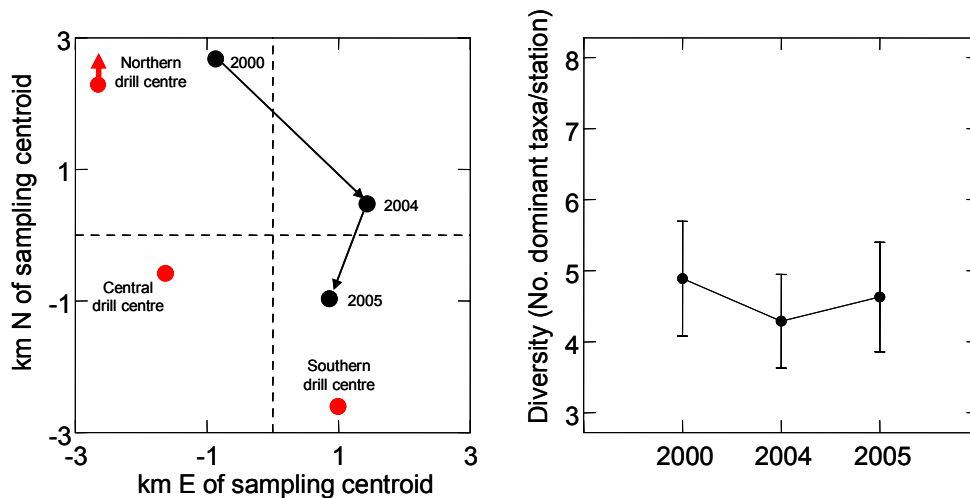


**Figure 5-45 Diversity versus Depth and Distance from the Northern and Central Drill Centres (2000, 2004, 2005)**



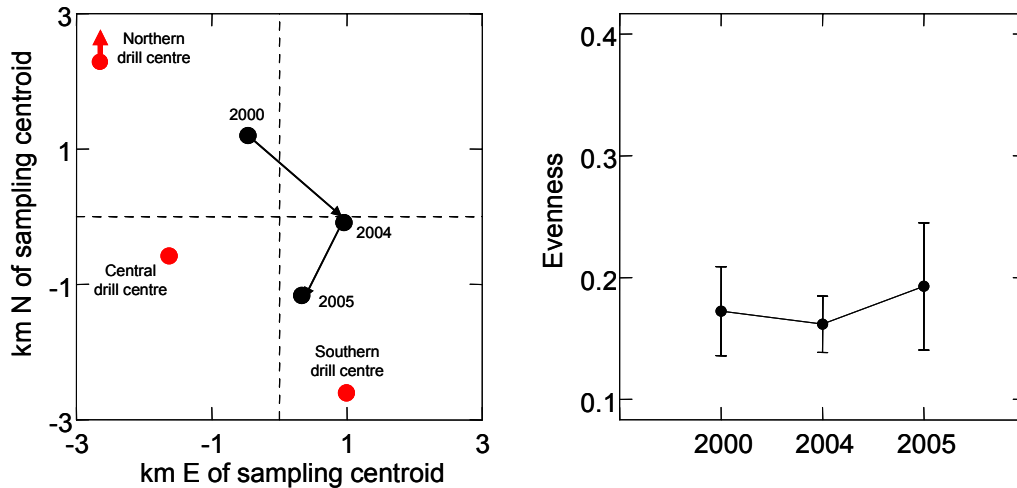
**Figure 5-46 Richness Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values



**Figure 5-47 Diversity Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

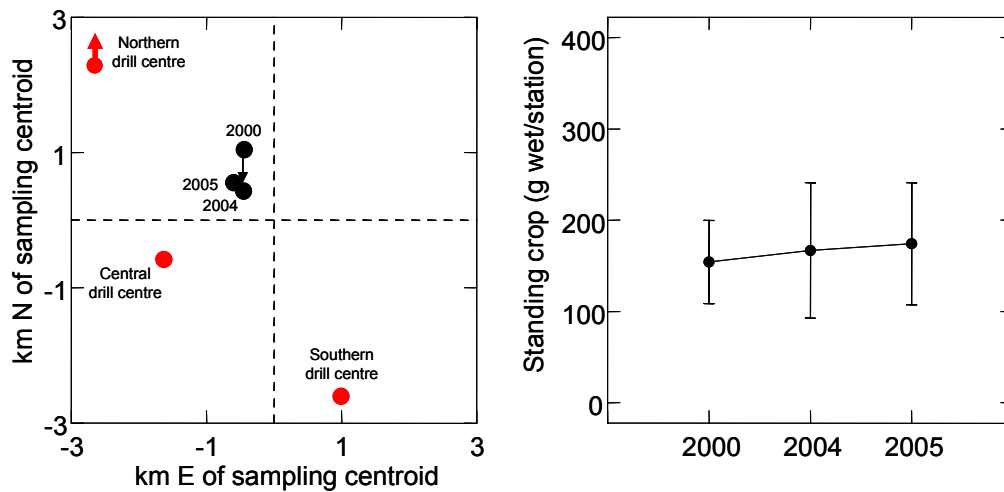


**Figure 5-48 Evenness Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

**Standing Crop and MDS2**

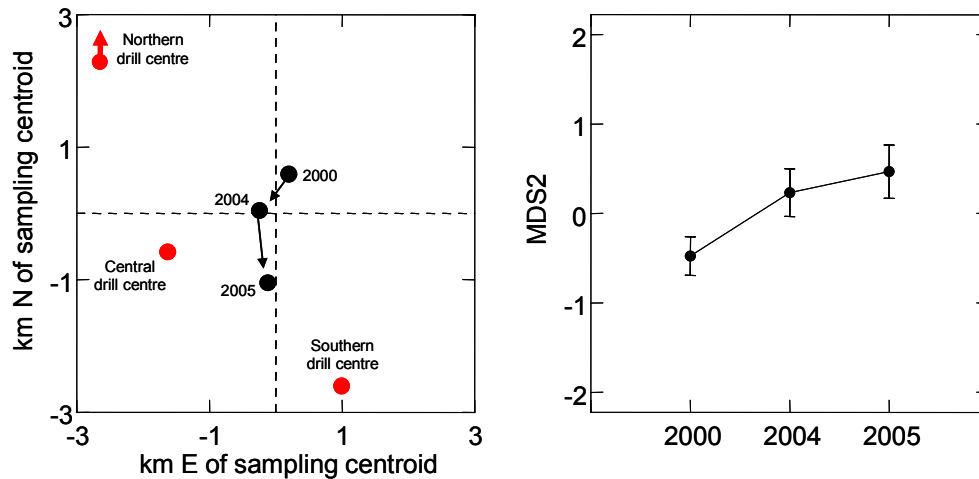
The only significant term for standing crop in the RM regressions analysis was Among Stations Error 1 or carry-over effects (Table 5-29). Standing crop has always been somewhat greater to the north and west of the sampling centroid, and has not changed significantly over time (Figure 5-49).



**Figure 5-49 Standing Crop Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

None of the terms in Table 5-29 was significant for MDS2. The lack of significance for the Within Stations Year terms was surprising, since MDS2 scores have increased over time as some taxa have replaced others (Figure 5-50; see also Table 5-20 and Figure 5-31). However, these changes over time have not been consistent at the family level (i.e., Family A did not always replace Family B; instead, Family C may have replaced D in one year and Family E may have replaced Family F in the next year).



**Figure 5-50 MDS2 Centroids and Changes Over Time for the 37 Stations Sampled in 2000, 2004 and 2005**

Note: - Means and SD are based on the Y scale used. The Y axes include the full range of individual values

**Carry-over Effects**

Results for RM comparisons of benthic invertebrate community variables among the 42 stations sampled in 2004 and 2005 (stations 4 and 19 excluded) are not presented because they were similar to results for comparisons of the 37 stations sampled in all three years. However, as was the case for sediment physical and chemical characteristics (Section 5.4.1.3), carry-over effects, or persistent differences among stations unrelated to depth or distances from drill centres (Among Stations Error 1 effects), were much stronger in the comparison of 2004 versus 2005 than in the comparison of all three years (Table 5-31). For amphipod abundance and probably total abundance and MDS1, some of the carry-over effects may represent persistent localized or small-scale drilling effects unrelated to distance from the drill centres. However, for other variables, the appropriate conclusion is that natural small-scale spatial differences persist over the short term but not the long term.

**Table 5-31 Carry-over Effects for Benthic Invertebrate Community Variables (2000, 2004, 2005)**

Benthic invertebrate community variable	2000, 2004 and 2005 (n=37 stations)		2004 versus 2005 (n=42 stations)	
	F	p	F	p
Total abundance	1.25	0.221	<b>3.11</b>	<b>&lt;0.001</b>
Standing crop	1.64	0.047	<b>2.50</b>	<b>&lt;0.001</b>
Richness	1.06	0.415	2.25	0.003
Diversity	0.65	0.907	<b>4.94</b>	<b>&lt;0.001</b>
Evenness	0.56	0.961	<b>2.77</b>	<b>&lt;0.001</b>
MDS1	1.46	0.100	<b>2.69</b>	<b>&lt;0.001</b>
MDS2	0.96	0.542	2.24	0.003
No. Amphipoda	1.49	0.089	<b>3.03</b>	<b>&lt;0.001</b>

Notes: - Carry-over effects are persistent differences among stations unrelated to depth or distance (Among Stations Error in RM models)  
 - Effects significant at  $p \leq 0.001$  in bold

**5.4.4 Integrated Assessment**

**5.4.4.1 Relationships Between Benthic Communities and Sediment Particle Size and TOC Content**

Table 5-32 provides rank correlations ( $r_s$ ) between benthic invertebrate community variables and sediment fines, gravel and TOC for all stations sampled in each sample year (2000, 2004 and 2005) and results of van Belle tests comparing correlations among years and testing average correlations over all three years. Depth correlations were added, because depth effects were significant for some community variables.

**Table 5-32 Spearman Rank Correlations ( $r_s$ ) Between Benthic Invertebrate Community Variables versus Sediment Particle Size and TOC and Depth (2000, 2004 and 2005)**

X variable	Y variable	$r_s$			Differences in $r_s$ among years	Mean $r_s$
		2000 (n=46 stations)	2004 (n=56 stations)	2005 (n=44 stations)		
% fines	Abundance	0.053	0.117	-0.150	NS	0.012
	Standing crop	0.031	-0.198	-0.123	NS	-0.100
	Richness	0.195	0.274*	-0.151	NS	0.114
	Diversity	0.153	0.230	0.101	NS	0.164
	Evenness	-0.015	0.008	0.145	NS	0.044
	MDS1	-0.097	-0.236	-0.216	NS	-0.185*
	MDS2	0.076	0.001	-0.039	NS	0.013
% gravel	No. Amphipoda	0.051	-0.055	-0.051	NS	-0.019
	Abundance	0.221	-0.189	0.084	NS	0.031
	Standing crop	-0.170	0.048	-0.139	NS	-0.082
	Richness	0.288	0.255	0.118	NS	0.222**
	Diversity	0.198	0.292*	0.304*	NS	0.266**
	Evenness	0.064	0.085	-0.029	NS	0.042
	MDS1	-0.245	-0.175	-0.213	NS	-0.210*
	MDS2	-0.012	-0.191	0.066	NS	-0.052
No. Amphipoda	0.284	0.067	-0.056	NS	0.098	

X variable	Y variable	$r_s$			Differences in $r_s$ among years	Mean $r_s$
		2000 ( $n=46$ stations)	2004 ( $n=56$ stations)	2005 ( $n=44$ stations)		
TOC	Abundance	-0.145	-0.122	-0.075	NS	-0.115
	Standing crop	0.156	0.025	-0.166	NS	0.007
	Richness	-0.136	0.071	-0.080	NS	-0.044
	Diversity	0.167	0.162	0.060	NS	0.131
	Evenness	0.144	0.170	0.139	NS	0.152
	MDS1	-0.130	-0.230	0.000	NS	-0.125
	MDS2	0.090	0.122	-0.288	NS	-0.018
	No. Amphipoda	0.042	-0.179	0.249	NS	0.028
Depth	Abundance	0.382*	0.171	0.286	NS	<b>0.276***</b>
	Standing crop	0.025	0.011	-0.117	NS	-0.025
	Richness	0.260	0.331*	0.207	NS	0.269**
	Diversity	0.043	0.380**	0.339*	NS	0.258**
	Evenness	-0.160	0.147	0.025	NS	0.009
	MDS1	-0.080	-0.438**	-0.037	NS	-0.195*
	MDS2	0.004	-0.172	-0.186	NS	-0.120
	No. Amphipoda	-0.045	0.108	0.239	NS	0.100

Notes: - NS—Not Significant ( $p > 0.05$ ); \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - Differences among  $r_s$  and mean  $r_s$  were tested using the van Belle test (Appendix B-5)  
 - Richness, diversity, evenness and MDS scores were based on families

Correlations between community variables and sediment particle size within years were weak, rarely significant at  $p \leq 0.05$ , and never significant at  $p \leq 0.01$  (Table 5-32). No within-year correlations for TOC were significant at  $p \leq 0.05$ . Differences among years were never significant at  $p \leq 0.05$ , partly because most correlations were weak but also partly because the van Belle test for differences is not powerful with only three blocks (i.e., years). Several mean correlations for particle size were significant, some at  $p < 0.01$ . Tests of mean correlations were powerful because the effective sample size was 156 stations. Correlations with depth were stronger for total abundance, richness, diversity and MDS1, consistent with parametric relationships identified in Section 5.4.3.4. The extreme depths at stations 4 and 19 (sampled 2004 and 2005) had limited effects on depth correlations after ranking. It was surprising that correlations with fines content were not stronger for these four invertebrate community variables, since fines content increased significantly with depth (Section 5.4.1.3).

Richness and diversity were positively correlated, and MDS1 (polychaete dominance) negatively correlated, with gravel content over all three years, and these were the strongest correlations (mean  $r_s$ ) with particle size in Table 5-32. The three community variables were also significantly positively correlated with depth, and both fines and gravel content tended to increase with increasing depth. To separate the effects of particle size (including fines) and depth (X), rank-rank regressions for the three community (Y) variables were compared among years in ANCOVA (Table 5-33). Both gravel and depth effects were significant for richness and diversity, and stronger (more significant) for diversity. This could be evidence of a habitat heterogeneity effect, with habitat diversity (variance of interstitial space sizes) increasing with increasing gravel content. As in the analysis of fines content alone (Table 5-32), fines effects were not significant.

**Table 5-33 Results of ANCOVA Assessing Particle Size and Depth Effects on Selected Benthic Invertebrate Community Variables (2000, 2004 and 2005)**

Y variable	X variable		
	% fines	% gravel	Depth
Richness	NS	*	*
Diversity	NS	***	**
MDS1	NS	*	NS

Notes: - NS—Not Significant ( $p > 0.05$ ); \* $p \leq 0.05$ ; \*\* $p \leq 0.01$ ; \*\*\* $p \leq 0.001$  (in bold)  
 - The Year term tests for differences in Y values among years across the entire range of X values  
 - Richness, diversity and MDS scores were based on families

Results for MDS1 were more equivocal. Depth effects were not quite significant at  $p \leq 0.05$  ( $p = 0.073$ ) whereas gravel effects were ( $p = 0.015$ ). Effects of the two X variables should be considered similar in strength, since whether  $p$  for gravel and depth effects are above or below 0.05 depends on the data set analyzed and the model/transformation used for analysis. The important point is that apparent depth effects on MDS1, which were significant in the RM analysis (Section 5.4.3.4), may have been partially attributable to covariance of gravel and depth.

**5.4.4.2 Relationships Between Benthic Communities and Tracers (Barium, >C<sub>10</sub>-C<sub>21</sub> HCs)**

Table 5-35 provides rank correlations ( $r_s$ ) between benthic invertebrate community variables and drilling tracers (barium, >C<sub>10</sub>-C<sub>21</sub> HCs) for all stations sampled in each sample year (2000, 2004 and 2005), and results of van Belle tests comparing correlations among years and testing average correlations over all three years. Baseline (2000) correlations between community variables and >C<sub>10</sub>-C<sub>21</sub> HCs were considered to be 0, since all >C<sub>10</sub>-C<sub>21</sub> HC concentrations were below EQL. Baseline correlations for barium should be considered estimates of natural relationships within a relatively narrow range of concentrations (120 to 210 mg/kg). Within years, many correlations with tracers were significant, including some baseline (2000) correlations with barium. Despite the limited power of the test for differences in correlations among years, these differences were often significant. Mean correlations were not calculated when differences among years were significant.

**Table 5-34 Spearman Rank Correlations ( $r_s$ ) Between Benthic Invertebrate Community Variables versus Barium and >C<sub>10</sub>-C<sub>21</sub> HCs (2000, 2004 and 2005)**

X variable	Y variable	$r_s$			Differences in $r_s$ among years	Mean $r_s$
		2000 (n=46 stations)	2004 (n=56 stations)	2005 (n=44 stations)		
Barium	Abundance	0.091	-0.167	-0.473**	*	
	Standing crop	-0.213	-0.017	-0.048	NS	-0.090
	Richness	0.417**	0.183	-0.316*	**	
	Diversity	0.428**	0.175	0.113	NS	0.238**
	Evenness	0.107	0.057	0.350*	NS	0.166*
	MDS1	-0.443**	-0.354**	<b>-0.579***</b>	NS	<b>-0.454***</b>
	MDS2	<b>-0.523***</b>	-0.210	0.305*	***	
	Stn. 4, 19 excl.	<b>-0.523***</b>	-0.217	0.307*	***	
No. Amphipoda	0.329*	-0.419**	<b>-0.573***</b>	***		

X variable	Y variable	$r_s$			Differences in $r_s$ among years	Mean $r_s$
		2000 (n=46 stations)	2004 (n=56 stations)	2005 (n=44 stations)		
>C <sub>10</sub> -C <sub>21</sub> HCs	Abundance	0.000	-0.170	<b>-0.534***</b>	*	
	Standing crop	0.000	-0.006	-0.075	NS	-0.026
	Richness	0.000	-0.140	-0.313*	NS	-0.150
	Diversity	0.000	-0.121	-0.087	NS	-0.071
	Evenness	0.000	-0.025	0.272	NS	0.077
	MDS1	0.000	0.119	<b>-0.515***</b>	**	
	MDS2	0.000	0.109	0.419**	NS	0.172*
	Stn. 4, 19 excl.	0.000	0.026	0.352*	NS	0.120
No. Amphipoda	0.000	-0.440**	<b>-0.729***</b>	**		

Notes: - NS—Not Significant ( $p > 0.05$ ); \*— $p \leq 0.05$ ; \*\*— $p \leq 0.01$ ; \*\*\*— $p \leq 0.001$  (in bold)  
 - Mean  $r_s$  were not calculated when  $p \leq 0.05$  for test of equality of  $r_s$  among years  
 - Richness, diversity, evenness and MDS scores were based on families

The emphasis below is on relationships with >C<sub>10</sub>-C<sub>21</sub> HCs, which was a superior tracer and predictor of biological effects on invertebrate communities. Barium was an effective tracer of drilling activity and would be the only available tracer if SBMs were not used. However, barium has serious deficiencies as a correlate or predictor (i.e., X variable) of biological effects in parametric analyses. The fundamental problem was that variance in invertebrate community (Y) variables was wide within the narrow background range of barium concentrations (120 to 210 mg/kg) measured in 2000, and most post-drilling concentrations were within that range. Alterations attributable to drilling could only be clearly identified at barium concentrations greater than 200 - 250 mg/kg, or for about 5 or 10 stations in each of 2004 and 2005 (i.e., there were only 5 or 10 “useful” X values for effects assessment). Even then, apparent natural (2000) relationships over a narrow range of barium concentrations could be as strong as relationships that also included apparent effects over a broader range of concentrations in 2004 and 2005 (e.g., compare correlations between MDS1 and barium in Table 5-34 over time). Appendix B-5 provides further results and discussion of relationships between invertebrate community variables and barium concentrations.

For sediment, any concentration of >C<sub>10</sub>-C<sub>21</sub> HCs greater than EQL (0.3 mg/kg) can be considered evidence of contamination from drilling. Baseline (2000) and remote/reference concentrations in 2004 and 2005 were below EQL. Consequently, >C<sub>10</sub>-C<sub>21</sub> HCs provided a broader and more continuous range of X values outside the range of background values for effects assessment. In other words, there were more than 40 quantitative and useful values or stations in 2004 and 2005 for effects assessment (and 46 useful values in 2000 for convincingly defining background levels). Appendix B-5 indicates that there were few or no drilling/tracer effects evident from analysis of barium that were not more evident or stronger based on analyses of >C<sub>10</sub>-C<sub>21</sub> HCs.

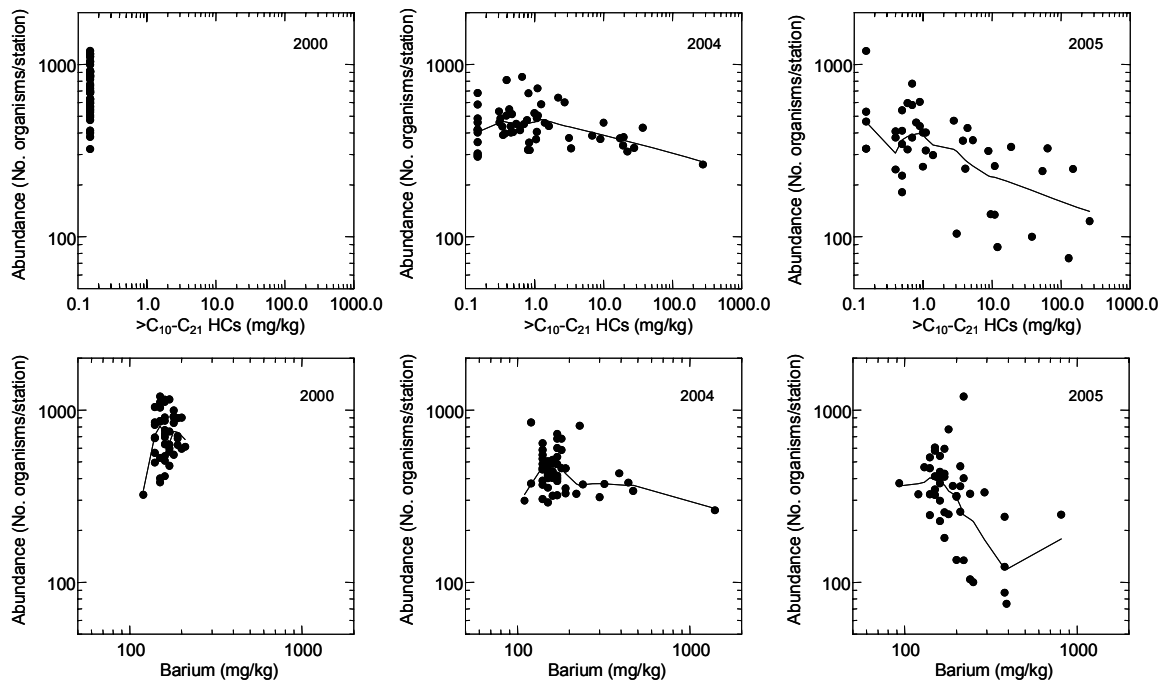
**Total Abundance, MDS1 and Amphipod Abundance**

In 2005, correlations between total abundance, MDS1 and amphipod abundance and >C<sub>10</sub>-C<sub>21</sub> HCs were negative (i.e., decreases in community variable values with increasing concentrations) and significant at  $p \leq 0.001$  (Table 5-34). These correlations were weaker in 2004, suggesting that drilling effects became stronger or only occurred in



2005, consistent with results for analyses of distance (Section 5.4.3.4). For all three variables, differences in correlations with  $>C_{10}\text{-}C_{21}$  HC among years were significant.

Figure 5-51 plots relationships between total abundance and tracer concentrations. Baseline (2000) data were included to illustrate the natural range and variance of Y values. The lines in the plots are LOWESS (Locally Weighted Scatter-plot Smoothers) trend lines (see B-5 for details), and were included to suggest possible parametric regression models. Plots of total abundance versus barium were included to indicate that, in 2004 and 2005, relationships at higher barium concentrations (i.e., greater than 200-250 mg/kg) were similar to those for  $>C_{10}\text{-}C_{21}$  HCs, but that variance in total abundance at lower barium concentrations within the narrow background range (less than 250 mg/kg) was also high (especially in 2004).



**Figure 5-51 Total Abundance versus Barium and  $>C_{10}\text{-}C_{21}$  HC Concentrations (2000, 2004 and 2005)**

In 2004, total abundance may have decreased with increasing  $>C_{10}\text{-}C_{21}$  HC concentrations at intermediate to high levels (Figure 5-51), but overall rank correlations were not significant (Table 5-34). In 2005, abundance decreased significantly with increasing  $>C_{10}\text{-}C_{21}$  HC concentrations. For 2004, with  $>C_{10}\text{-}C_{21}$  HCs as X, a hockey-stick model significantly reduced error variance relative to a bivariate regression, although *R* was low (Table 5-35). The estimated threshold concentration (i.e., the concentration above which effects occurred) was 2 mg/kg, but the 95% CI of 0.2 (<EQL) to 30.9 mg/kg included approximately half the range of observed concentrations. In contrast, for 2005, the relationship between total abundance and  $>C_{10}\text{-}C_{21}$  HCs was linear throughout the entire range of observed concentrations (i.e., the relationship was a shaft with no blade). A threshold within the range of observed concentrations could not be estimated. The intercept and slope for the 2005 bivariate regression are provided in Table 5-36.

**Table 5-35 Results for Parametric Dose-Response Models for Total Abundance, MDS1 and Amphipod Abundance versus >C<sub>10</sub>-C<sub>21</sub> HC Concentrations (2004, 2005)**

Year	Result/Estimate	Total abundance	MDS1	No. Amphipoda
2004	Bivariate <i>r</i>	0.285*	0.020	<b>0.661***</b>
	Hockey-stick <i>R</i>	0.400**	0.365*	<b>0.739***</b>
	<i>p</i> threshold	0.030	0.006	<b>&lt;0.001</b>
	X <sub>T</sub> (threshold in mg/kg)	2.2	37.0	4.9
	95% CI	0.2 to 30.9	9.2 to 149.4	2.2 to 10.8
2005	Bivariate <i>r</i>	<b>0.618***</b>	<b>0.623***</b>	<b>0.773***</b>
	Hockey-stick <i>R</i>	<b>0.618***</b>	<b>0.649***</b>	<b>0.773***</b>
	<i>p</i> threshold	1.000	0.130	1.000
	X <sub>T</sub> (threshold in mg/kg)	None	0.9	None
	95% CI		0.2 to 4.6	

Notes: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)  
 - >C<sub>10</sub>-C<sub>21</sub> HC concentrations, total abundance and amphipod abundance (+ 1) were log-transformed  
 - MDS1 scores were based on families

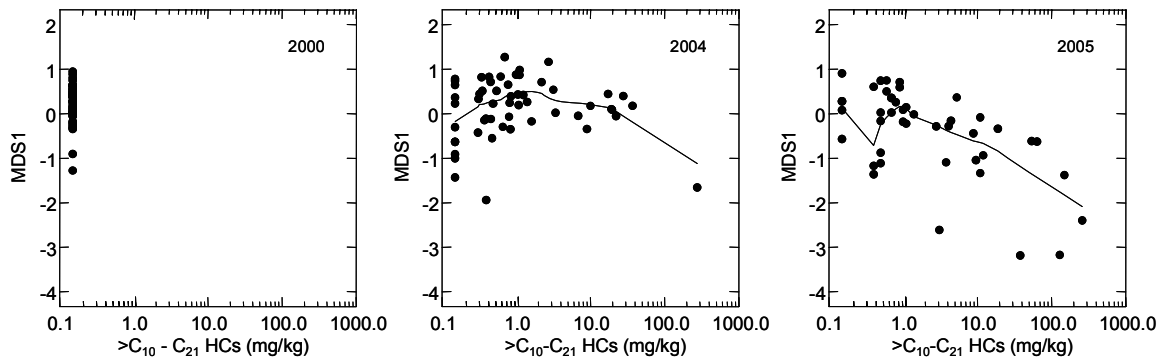
**Table 5-36 Intercepts and Slopes for Regressions of Total Abundance, MDS1 and Amphipod Abundance on >C<sub>10</sub>-C<sub>21</sub> HC Concentrations (2005)**

Y variable	Intercept (a) ± SE	Slope (b) ± SE
Total abundance (log <sub>10</sub> )	2.545 ± 0.032	-0.180 ± 0.035
MDS1	-0.199 ± 0.127	-0.711 ± 0.138
No. Amphipoda + 1 (log <sub>10</sub> )	0.971 ± 0.047	-0.403 ± 0.051

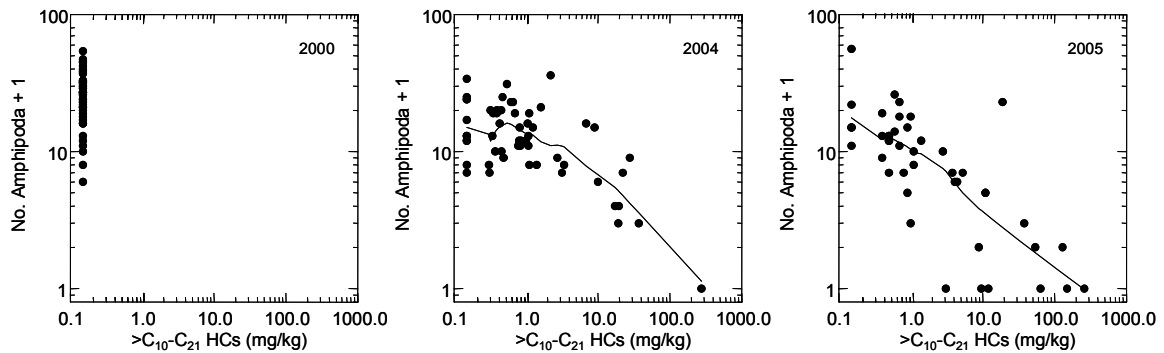
Notes: - \**p* ≤ 0.05; \*\**p* ≤ 0.01; \*\*\**p* ≤ 0.001 (in bold)  
 - >C<sub>10</sub>-C<sub>21</sub> HC concentrations, total abundance and amphipod abundance (+ 1) were log-transformed  
 - MDS1 scores were based on families

There was no correlation between MDS1 scores and >C<sub>10</sub>-C<sub>21</sub> HCs in 2004, but MDS1 scores decreased significantly with increasing >C<sub>10</sub>-C<sub>21</sub> HC concentrations in 2005 (Table 5-34; Figure 5-52). A significant threshold relationship could be fit to the 2004 data, but the estimated threshold was 37 mg/kg, towards the upper end of the observed concentration range and with wide CI (Table 5-35). In contrast, for 2005, a threshold model did not significantly reduce error relative to a bivariate model, the estimated threshold concentration of 0.9 mg/kg was close to EQL (0.3 mg/kg), and the lower 95% CL for the threshold was less than EQL. Table 5-36 provides the intercept and slope for the 2005 bivariate regression.

Amphipod abundance significantly decreased with increasing >C<sub>10</sub>-C<sub>21</sub> HC concentrations in both 2004 and 2005, with the correlation/relationship stronger in 2005 (Table 5-34; Figure 5-53). For 2004, the threshold concentration was 4.9 mg/kg; for 2005, the relationship was linear throughout the entire concentration range (Table 5-35). Table 5-36 provides the intercept and slope for the 2005 bivariate relationship.



**Figure 5-52 MDS1 versus >C<sub>10</sub>-C<sub>21</sub> HC Concentrations (2000, 2004 and 2005)**



**Figure 5-53 Amphipod Abundance versus >C<sub>10</sub>-C<sub>21</sub> HC Concentrations (2000, 2004 and 2005)**

The dose-response relationships for total abundance, MDS1 and amphipod abundances versus >C<sub>10</sub>-C<sub>21</sub> HCs were useful additions to the distance regressions provided in Section 5.4.3. Overall, the two approaches were in good agreement (Table 5-37), indicating that:

- effects on total abundance and MDS1 were relatively weak in 2004, but increased substantially in strength in 2005; and
- effects on amphipods were evident in 2004, but also increased in strength in 2005

The agreement was expected; >C<sub>10</sub>-C<sub>21</sub> HC concentrations were effectively the inverse of distance, given the strong negative correlation between the two X variables.

**Table 5-37 Comparison of Parametric Regressions with Total Abundance, MDS1 and Amphipod Abundance as Y and Distance versus >C<sub>10</sub>-C<sub>21</sub> HCs as X (2004 and 2005)**

Y variable	Year	Model	Distance	Fuel
Total abundance	2004	Bivariate <i>r</i>	0.322*	0.285*
		Threshold <i>R</i>	0.340*	0.400**
		<i>p</i> threshold	0.395	0.030
	2005	Bivariate <i>r</i>	<b>0.520***</b>	<b>0.618***</b>
		Threshold <i>R</i>	<b>0.556***</b>	<b>0.618***</b>
		<i>p</i> threshold	0.136	1.000
MDS1	2004	Bivariate <i>r</i>	0.172	0.020
		Threshold <i>R</i>	0.324	0.365*
		<i>p</i> threshold	0.039	0.006
	2005	Bivariate <i>r</i>	<b>0.486***</b>	<b>0.623***</b>
		Threshold <i>R</i>	<b>0.617***</b>	<b>0.649***</b>
		<i>p</i> threshold	0.004	0.130
No. Amphipoda	2004	Bivariate <i>r</i>	<b>0.517***</b>	<b>0.661***</b>
		Threshold <i>R</i>	<b>0.618***</b>	<b>0.739***</b>
		<i>p</i> threshold	0.003	<0.001
	2005	Bivariate <i>r</i>	<b>0.655***</b>	<b>0.773***</b>
		Threshold <i>R</i>	<b>0.668***</b>	<b>0.773***</b>
		<i>p</i> threshold	0.268	1.000

>C<sub>10</sub>-C<sub>21</sub> HCs were a better predictor of community variable values than distance (i.e., *R* or *r* were higher) for most if not all direct comparisons (i.e., within rows) of the two *X* variables in Table 5-37. The differences between the two predictors were probably attributable to differences in distance effects from different drill centres plus directional and other effects that were not incorporated into the simple “distance from the nearest active drill centre” measure used, but were indirectly incorporated into variance of >C<sub>10</sub>-C<sub>21</sub> HC concentrations. These additional effects are mostly localized and would be most important near drill centres, where the wide range of >C<sub>10</sub>-C<sub>21</sub> HC concentrations would probably be a better predictor of biological effects than the narrow range of distances.

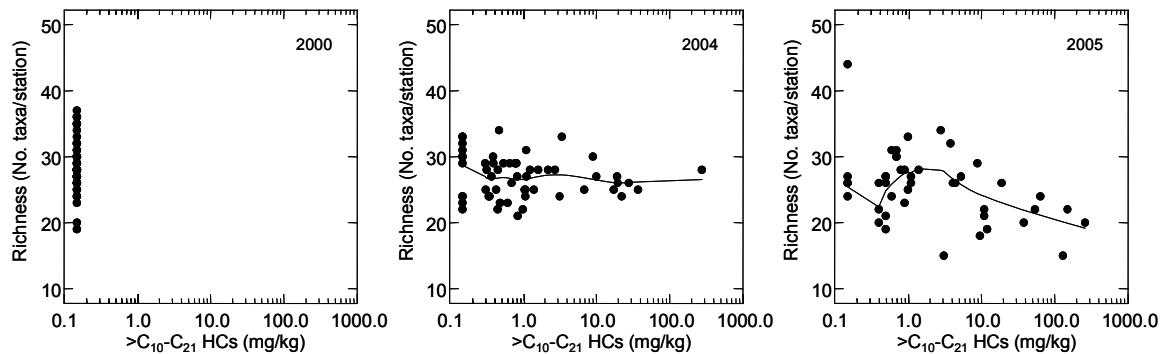
However, distance regressions are required to define zones of effects, if they exist. Distance regressions are also probably better predictors of biological effects for more remote stations (e.g., more than 10 or even 5 km from drill centres), where variance of >C<sub>10</sub>-C<sub>21</sub> HC concentrations near or below EQL was mostly analytical rather than attributable to variance in contamination from drilling.

### Richness and MDS2

Rank correlations between richness and MDS2 and >C<sub>10</sub>-C<sub>21</sub> HCs were 0 in 2000, near 0 in 2004, and increased in strength and became significant in 2005 (Table 5-34). Baseline (2000) correlations with barium also reversed over time for the two variables. These changes were consistent with hypotheses of the presence or intensification of drilling effects, undetected in analyses of distance (Section 5.4.3.4).

In 2004, there was no relationship between richness and >C<sub>10</sub>-C<sub>21</sub> HCs (Table 5-34; Figure 5-54). In 2005, richness decreased with increasing >C<sub>10</sub>-C<sub>21</sub> HC concentration over a relatively broad range (i.e., beyond 1 to 5 mg/kg), and the overall rank correlation between richness and concentration was significant, although weak. However, any relationship between richness and >C<sub>10</sub>-C<sub>21</sub> HCs in 2005 was almost entirely a function of effects on total abundance, which was strongly positively correlated with richness (Table 5-22). With total abundance included as an additional *X* variable in parametric or

non-parametric regression/correlation analyses, effects of  $>C_{10}-C_{21}$  HCs on richness were not significant. In other words, there were no additional effects on richness beyond what would be expected from reductions in abundance. Note that diversity, which also adjusts richness for variance in abundance, was uncorrelated with  $>C_{10}-C_{21}$  HCs (Table 5-34).



**Figure 5-54 Richness versus  $>C_{10}-C_{21}$  HC Concentrations (2000, 2004 and 2005)**

MDS2 scores were uncorrelated or weakly positively correlated with  $>C_{10}-C_{21}$  HC concentrations in 2004, but increased significantly with  $>C_{10}-C_{21}$  HC concentrations in 2005 (Table 5-34). The low MDS2 values at reference stations 4 and 19 (not sampled in 2000) in 2004 and 2005 were clearly outliers (Figure 5-55, top row), and  $>C_{10}-C_{21}$  HC concentrations were below EQL at both stations in both years. However, these two stations had minimal influence on LOWESS trend lines and rank correlations with  $>C_{10}-C_{21}$  HC concentrations. Based on the LOWESS trend lines with stations 4 and 19 deleted (Figure 5-55, bottom row), it was surprising that there was any difference in correlations with  $>C_{10}-C_{21}$  HCs between 2004 and 2005, and a significant increase in MDS2 with increasing concentration in 2005. All that can be concluded is that no effects-based (or depth-based) model could ever predict:

- the increase in MDS2 scores between 2000 versus 2004 and 2005 at all or most stations; and
- the unusually low MDS2 scores at stations 4 and 19 in 2004 and 2005, even relative to 2000 values.

MDS2 was a secondary axis of variance in community composition reflecting differences in the abundances of a broad range of minor taxa. With variance in the major axis of community composition and abundances of dominant taxa (i.e., MDS1) related to distance and  $>C_{10}-C_{21}$  (i.e., drilling) and depth, it would be reasonable to assume that variance in MDS2 scores over space and time was primarily related to other factors.

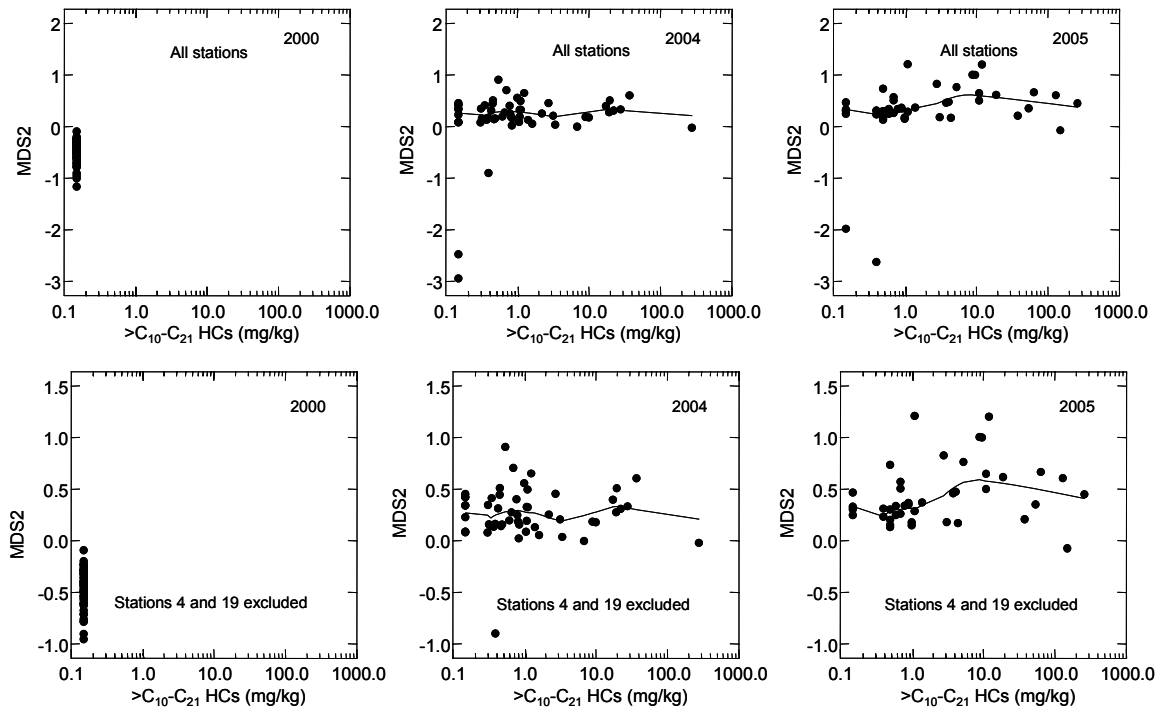


Figure 5-55 MDS2 versus >C<sub>10</sub>-C<sub>21</sub> HC Concentrations (2000, 2004 and 2005)

## 5.5 Summary of Findings

### 5.5.1 Physical and Chemical Characteristics

Sediments collected from 44 stations in 2005 were predominantly (97.4%) sand. Fines (1.2%) and TOC content (0.89 g/kg or 0.089%) were low.

PAHs, benzene, ethylbenzene and xylenes were not detected at any station in 2005 at an EQL of 0.03 mg/kg. Toluene was detected at one station at 0.04 mg/kg (EQL: 0.03 mg/kg). >C<sub>10</sub>-C<sub>21</sub> HCs were detected at 39 of 44 stations at an EQL of 0.3 mg/kg. >C<sub>21</sub>-C<sub>32</sub> HCs were detected at 19 of 44 stations at an EQL of 0.3 mg/kg.

Aluminum, barium, chromium, iron, lead, manganese, strontium, uranium, vanadium, zinc and ammonia were detected at all 44 stations. Sulphur was detected at 43 stations at an EQL of 0.02%.

In 2005, fines and TOC content were significantly correlated. Concentrations of metals other than barium, and ammonia, were significantly positively correlated with fines and TOC content. Barium and >C<sub>10</sub>-C<sub>21</sub> HC concentrations, used as tracers of drilling muds, were strongly positively correlated. Sulphur concentrations were significantly positively correlated, and redox levels were significantly negatively correlated, with concentrations of the two drilling mud tracers. Barium concentrations were also weakly but significantly positively correlated with concentrations of other metals, a natural relationship also observed during baseline (2000) sampling. Concentrations of metals other than barium were uncorrelated with >C<sub>10</sub>-C<sub>21</sub> HC concentrations. Ammonia concentrations were uncorrelated with concentrations of tracers, metals, sulphur and redox levels.

In 2005, concentrations of barium and  $>C_{10}-C_{21}$  HCs decreased significantly with distances from drill centres. Estimated zones of influence were 2.6 km (95% CI: 2.1 to 6.3 km) for barium and 6.5 km (95% CI: 3.9 to 11.8 km) for  $>C_{10}-C_{21}$  HCs. These zones of influence were based on distance from the nearest drill centre (Northern, Central, Southern), since using distances from each drill centre did not improve distance regression models and predictions. Concentrations of the two tracers were generally greater to the southeast of the Central and/or Southern drill centres, in the direction of residual currents.

Relationships between barium concentrations and distance from the Northern drill centre did not change between baseline (2000) and EEM years (2004, 2005), after drilling began at this centre. In contrast, relationships with distance from the Southern drill centre changed substantially and significantly between baseline and EEM years after drilling started, from “no relationship” to a “strong decrease in concentration with distance”. A similar change in relationships with distance from the Central drill centre occurred between 2004 and 2005, after drilling began at the Central drill centre. Overall barium concentrations progressively increased over time.

In 2000, all  $>C_{10}-C_{21}$  HC concentrations were less than EQL (0.03 mg/kg). In 2004 and 2005, most concentrations were greater than EQL.  $>C_{10}-C_{21}$  HC concentrations decreased significantly with distances from the Northern and Southern drill centres in 2004 and 2005, after drilling started at these two centres. A similar decrease with distance from the Central drill centre was not observed until 2005, after drilling started at this centre. Overall  $>C_{10}-C_{21}$  HC concentrations progressively increased over time.

In 2005, redox levels increased with distance from the nearest drill centre, with an estimated zone of influence of 2.6 km (95% CI: 1.0 to 6.4 km). However, using distance from each drill centre improved distance regressions, and redox levels decreased rather than increased with distance from the Northern drill centre.

In 2004, sulphur concentrations decreased with distance from both the Northern and Southern drill centres. However, in 2005, the relationship between sulphur concentrations and distance from the Northern drill centre was reversed (i.e., concentrations increased with distance), the decrease with distance from the Southern drill centre was weaker, and there was no overall relationship between sulphur concentrations and distances from the drill centres. Sulphur concentrations were also unrelated to distance from the Central drill centre in both 2004 and 2005.

Fines content consistently and significantly increased with depth, and was unrelated to distance from the Central drill centre, in all three sample years (2000, 2004 and 2005). In 2004, fines content increased with distance from the Northern drill centre, but decreased with distance from the Southern drill centre. In 2005, there was no overall relationship between fines content and distances from the drill centres (however defined).

TOC content increased with distance from the Central drill centre, and was unrelated to depth and distances from the Northern and Southern drill centres, in all three sample years.

Concentrations of metals other than barium decreased with distance from the Southern drill centre in 2004, but not in 2000 and 2005. The 2004 distance relationship was largely

a function of reduced concentrations at remote stations rather than increased concentrations at stations near the Southern drill centre.

Ammonia concentrations were unrelated to distances from the Central and Southern drill centres in 2004. In 2005, ammonia concentrations increased with distance from the Central drill centre and decreased with distance from the Southern drill centre, and there was no overall relationship between ammonia concentrations and distances from the drill centres.

Carry-over effects, or persistent differences among stations unrelated to distance or depth, were small and generally not significant over all three sample years for sediment physical and chemical variables. However, when only 2004 and 2005 were compared, carry-over effects were much larger and usually significant.

### **5.5.2 Toxicity**

No sediment samples were toxic to bacteria in 2000, 2004 and 2005 when tested in laboratory toxicity tests.

No sediment samples were toxic to amphipods in 2000 and 2004 when tested in laboratory toxicity tests. In 2005, sediment from one station was toxic to amphipods (survival: 28%), and survival in sediment from another station (67.5%) was lower than in samples from other stations sampled in 2000, 2004 and 2005 (survival has usually been greater than 80%). These two stations with low survival were not extreme in terms of proximity to drill centres or sediment physical or chemical characteristics.

### **5.5.3 Benthic Community Structure**

In each sample year (2000, 2004 and 2005), polychaetes accounted for approximately 75% of the invertebrates collected. Bivalves accounted for approximately 17% of the total in each year. Amphipoda, Tanaidacea and Echinodermata were the only other "major" (higher-level) taxa accounting for more than 1% of total abundance in one or more years.

The primary patterns of variance in community composition were related to the relative abundances of the two dominant major taxa (i.e., polychaetes versus bivalves). When relative abundances of polychaetes increased, relative abundances of bivalves decreased, and vice versa. Three families, the polychaetes Spionidae and Paraonidae (polychaetes) and the bivalve Tellinidae, accounted for 65 to 70% of the invertebrates collected. Secondary patterns of variance over space and time were related to changes in abundances of sub-dominant polychaete, bivalve and amphipod families.

Total abundance was generally greater where and when polychaetes, the most abundant taxon, were dominant. Richness was positively correlated with total abundance, with more taxa (families) generally collected where and when more organisms, especially polychaetes, were collected. Diversity was largely unrelated to total abundance.

In 2005, most benthic invertebrate community variables were not significantly correlated with sediment particle size, TOC content, concentrations of metals other than barium, or ammonia. Correlations between invertebrate community variables and drilling mud



tracers (barium, >C<sub>10</sub>-C<sub>21</sub> HCs) and redox were much stronger. Total abundance, polychaete dominance (i.e., polychaetes:bivalves), and amphipod abundance decreased with increasing tracer concentrations and decreasing redox levels.

In 2005, total abundance, polychaete dominance and amphipod abundance significantly increased with increasing distance from the drill centres. Reductions in total abundance and polychaete dominance were primarily a function of effects of the Southern drill centre; reductions in amphipod abundance were a function of effects from all three drill centres (see below). The estimated zone of effects for reductions in total abundance was 2.6 km (95% CI: 1 to 6 km) from the nearest drill centre. This estimate may be suspect because total abundances also increased with increasing depth in all three years. The estimated zone of effects for reductions in polychaete dominance was 2.2 km (95% CI: 1.1 to 4.2 km). The increases in amphipod abundance with increasing distance from the drill centres extended over most stations and distances (i.e., a zone of effects would include all or most of the stations sampled).

Distance effects on total abundance, polychaete dominance and amphipod abundance were considerably weaker in 2004 than in 2005. For the 37 stations sampled in 2000, 2004 and 2005, total abundance decreased with distance from the Southern drill centre in 2000. This baseline relationship was reversed in 2004 and 2005, with increases in abundance with distance significantly stronger in 2005 than in 2004. In all three sample years, there was no relationship between total abundance and distance from the Northern drill centre, and abundance decreased with distance from the Central drill centre.

In all three sample years, there was no relationship between polychaete dominance and distances from the Northern and Central drill centres. The strong increase in polychaete dominance with distance from the Southern drill centre observed in 2005 was not evident in 2000 and 2004.

In 2000, amphipod abundance decreased with distances from all three drill centres. The baseline distance gradients for the Northern and Southern drill centres were reversed in 2004, after drilling began at these two centres. The baseline distance gradient for the Central drill centre was reversed in 2005, after drilling began at this centre.

Richness and diversity increased with increasing depth and decreased with distance from the Central drill centre, in all three sample years.

Carry-over effects for invertebrate community variables were weak and significant only for standing crop when all three sample years were compared. However, when only 2004 and 2005 were compared, carry-over effects were strong and significant (usually at  $p \leq 0.001$ ) for all variables.

#### **5.5.4 Integrated Assessment**

Both within years, and over all three sample years (2000, 2004 and 2005), benthic invertebrate community variables were uncorrelated with sediment fines and TOC content. Richness, diversity and polychaete dominance increased with increasing gravel content.

In 2004 and 2005, after drilling began, total abundance, polychaete dominance and amphipod abundance decreased with increasing tracer (barium and  $>C_{10}-C_{21}$  HC) concentrations. The decreases in community variable values with increasing barium concentrations were most apparent at concentrations greater than 250 mg/kg and outside the baseline or background range. These higher concentrations occurred at only 5 or 10 stations in each of 2004 and 2005. At most stations, community variables varied widely across a relatively narrow range of lower barium concentrations.

$>C_{10}-C_{21}$  HC concentrations in 2004 and 2005 were effective quantitative predictors, usually more effective than distances from drill centres, of post-drilling total abundance, polychaete dominance and amphipod abundance values. In 2004, estimated threshold concentrations (i.e., concentrations below which effects did not occur) were 2.2 mg/kg for total abundance, 37 mg/kg for polychaete dominance, and 4.9 mg/kg for amphipod abundance. In 2005, values of all three variables decreased with increasing concentration across all or most of the concentration range. Threshold concentrations could not be estimated for effects on total and amphipod abundances. The estimated threshold concentration for effects on polychaete dominance was 0.9 mg/kg. However, the 95% CI (0.2 to 4.6 mg/kg) for this threshold included the EQL of 0.3 mg/kg, and a threshold relationship was not a significant improvement over a simple bivariate regression of polychaete dominance on  $>C_{10}-C_{21}$  HC concentrations.

## 6.0 Commercial Fish Component

### 6.1 Field Collection

The *CCG Wilfred Templeman*, its crew and DFO Science personnel were chartered for the 2005 commercial fish survey of American plaice (“plaice”) and snow crab (“crab”) between July 8 and July 13, 2005. Collection dates for the baseline program and EEM programs, and tests performed on collected specimens, are shown in Table 6-1.

**Table 6-1 Field Trips Dates**

Trip	Collections/Tests	Date
2000 Baseline Program	Study Area Crab for Body Burden Analysis; Study and Reference Area plaice for body burden and taste analysis; Study Area plaice for health analysis.	July 4 to July 10, 2000
2002 Baseline Program	Reference Area crab for body burden analysis; Study and Reference Area crab for taste analysis; Reference Area plaice for health analysis.	June 24 to July 10, 2002
2004 EEM Program	Study and Reference Area plaice and crab for body burden and taste analysis. Study and Reference Area plaice for health analysis.	July 10 to July 18, 2004
2005 EEM Program	Study and Reference Area plaice and crab for body burden and taste analysis. Study and Reference Area plaice for health analysis.	July 8 to July 13, 2005

Notes: - Since the location of Reference Areas sampled in 2004 and 2005 differs from locations sampled in 2000 and 2002, data from Reference Areas collected during baseline can not be compared to EEM Reference Area data  
 - Study Area data are generally comparable

Details on the collection and processing of 2000, 2002 and 2004 samples are presented in Husky Energy (2001; 2003; 2005). Sampling for the 2005 program was conducted under a Department of Fisheries and Oceans Stock Assessment license. A total of 44 plaice (out of 148 fish caught) and 180 crab from the White Rose Study Area were retained for analysis in 2005. A total of 115 plaice (out of 244 caught) and 147 crab from the four Reference Areas were retained. Plaice that were not retained were released with as little damage as possible. Both plaice and crab were collected using a Campellan 1800 trawl towed at three knots for 15 minutes per transect. Because of limited time available for sampling, the liner was removed from the Campellan trawl in order to minimize by-catch and speed up sample processing time. Location of transects are provided in Figure 1-8 (Section 1) and Appendix C-1.

Preliminary processing of samples was done onboard ship. Plaice and crab that had suffered obvious trawl damage were discarded. Tissue samples, top fillet for plaice and left legs for crab, were frozen at -20°C for subsequent taste analysis. Bottom fillets and liver (left half only) for plaice and right legs for crab were frozen at -20°C for body burden analysis. Blood, gill, liver (right half), heart, spleen, gonad, kidney and otolith samples from plaice were preserved for fish health indicators analysis (see below). Additional measurements on plaice included fish length, weight (whole and gutted), sex and maturity stage, liver weight, and gonad weight. For crab, measurements included carapace width, shell condition (see Appendix C-1 for shell condition indices), sex and chela height. Only those plaice larger than 250 mm in length and those crab larger than 40 mm in carapace width were retained for analysis. This size cut-off for crab excluded female crab, which were smaller than 40 mm.

Plaice used in fish health analysis were killed by severing the spinal cord. Each fish was assessed visually for any parasites and/or abnormalities on the skin and fins. Blood was drawn from a dorsal vessel near the tail and dispensed carefully into a tube containing an anticoagulant (EDTA) and gently mixed. Two blood smears were then prepared for each fish within one hour of blood withdrawal according to standard haematological methods (Platt 1969). The entire liver was excised and bisected. A 4 to 5 mm thick slice was cut from the centre portion of the right half of the liver (along the longitudinal axis) and placed in 10% buffered formalin for histological processing and the rest was frozen on dry ice until return to port, when it was placed in a -65°C freezer for MFO analysis. The first gill arch on the right of the fish was removed and placed in 10% buffered formalin for histological processing. Tissue samples of heart, spleen and head-kidney were removed and placed in 10% buffered formalin for histological processing, if required. A pair of otoliths were removed for ageing. Throughout the dissection process, any internal parasites and/or abnormal tissues were recorded and preserved in 10% buffered formalin for subsequent identification.

Standard tissue sampling QA/QC protocols were followed for collection of samples to ensure sample integrity and prevent onboard contamination. The top deck of the survey vessel was washed with degreaser then flushed with seawater. The fishing deck and chute leading to the processing facilities were flushed continuously during the survey. Sampling personnel wore new latex gloves and all sampling and measuring instruments were washed with mild soap and water then rinsed with distilled water before each transect. Processed samples were transferred to a -20°C freezer within one hour of collection.

## **6.2 Laboratory Analysis**

### **6.2.1 Allocation of Samples**

Plaice from 11 trawls in the Study Area and 15 trawls in the Reference Areas were used for body burden analysis, taste tests and fish health. Plaice bottom fillets and half-livers were composited to generate 10 individual body burden samples for fillet and liver for the Study Area and three individual samples for each of the four Reference Areas. Fillet tissue from individual fish was archived for body burden on individuals if warranted by results of taste or health analyses. There was insufficient tissue to archive liver samples for individual fish. Top fillets from a subset of fish from each trawl used in body burden analysis were used in taste analysis. In this test, fish fillet selected from the Study Area and the Reference Areas were allocated to the triangle test and the hedonic scaling test (see Section 6.2.3 for details on taste tests) and randomly assigned to panelists. Fish health analyses focused on individual fish rather than composite or randomly assigned samples (Table 6-2).

**Table 6-2 Plaiice Selected for Body Burden, Taste and Health Analyses (2005)**

Transect Number	Group	Total No. Fish Retained	Body Burden Composites (Bottom Fillet, or Liver)	Taste (Number of Fish, Top Fillet)	Health (Number of Fish)
WR-01/05	Study	6	Composite 1 (6 fish)	2	6
WR-02	Study	6	Composite 2 (6 fish)	2	6
WR-03	Study	5	Composite 3 (5 fish)	2	5
WR-04	Study	6	Composite 4 (6 fish)	2	6
WR-06	Study	6	Composite 5 (6 fish)	2	6
WR-07	Study	5	Composite 6 (5 fish)	2	5
WR-08	Study	6	Composite 7 (6 fish)	2	6
WR-09	Study	6	Composite 8 (6 fish)	2	6
WR-10	Study	6	Composite 9 (6 fish)	2	6
WR-11	Study	8	Composite 10 (8 fish)	2	8
<b>Total</b>	<b>Study</b>	<b>60</b>	<b>10</b>	<b>20</b>	<b>60</b>
WR-13	Reference 2	10	Composite 11 (10 fish)	2	10
WR-14/16	Reference 2	10	Composite 12 (10 fish)	2	10
WR-15	Reference 2	10	Composite 13 (10 fish)	2	10
WR-17	Reference 1	10	Composite 14 (10 fish)	2	10
WR-18	Reference 1	10	Composite 15 (10 fish)	2	10
WR-19/20	Reference 1	10	Composite 16 (10 fish)	2	10
WR-21	Reference 4	10	Composite 17 (10 fish)	2	10
WR-22	Reference 4	10	Composite 18 (10 fish)	2	10
WR-23/24	Reference 4	10	Composite 19 (10 fish)	2	10
WR-25	Reference 1	10	Composite 20 (10 fish)	2	10
WR-26	Reference 1	10	Composite 21 (10 fish)	2	10
WR-27	Reference 1	10	Composite 22 (10 fish)	2	10
<b>Total</b>	<b>Reference</b>	<b>120</b>	<b>12</b>	<b>24</b>	<b>120</b>

Crab from 10 trawls in the Study Area and 13 trawls in the Reference Areas were used for body burden and taste analyses. All crab in each trawl, but excluding soft shell crab, were used. Tissue from right legs were composited to generate 10 individual body burden samples for the Study Area and one to three individual samples for each of the four Reference Areas (Table 6-3). Left leg tissue was used in taste analysis. In this test, leg tissue selected from the Study Area and the Reference Areas were allocated to the triangle test and the hedonic scaling test (see Section 6.2.3 for details on taste tests) and randomly assigned to panelists.

**Table 6-3 Crab Selected for Body Burden and Taste Analysis (2004)**

Transect Number	Group	Total No. of Crab	Body Burden Composites (Right Legs)	Taste (Number (wt) of Crab, Left Legs)
WR-01	Study	21	Composite 1 (21 crab)	3 (508g)
WR-02	Study	13	Composite 2 (13 crab)	6 (961g)
WR-03	Study	18	Composite 3 (18 crab)	8 (1024g)
WR-04	Study	16	Composite 4 (16 crab)	12 (1496g)
WR-06	Study	14	Composite 5 (14 crab)	8 (640g)
WR-07	Study	19	Composite 6 (19 crab)	6 (155g)
WR-08	Study	18	Composite 7 (18 crab)	6 (386g)
WR-09	Study	14	Composite 8 (14 crab)	6 (221g)
WR-10	Study	11	Composite 9 (11 crab)	11 (312g)
WR-11	Study	9	Composite 10 (9 crab)	6 (400g)
<b>Total</b>	<b>Study</b>	<b>153</b>		<b>72 (6103)</b>
WR-14	Reference 2	9	Composite 11 (9 crab)	6 (74g)
WR-15	Reference 2	6	Composite 12 (6 crab)	6 (112g)

Transect Number	Group	Total No. of Crab	Body Burden Composites (Right Legs)	Taste (Number (wt) of Crab, Left Legs)
WR-16	Reference 2	15	Composite 13 (15 crab)	12 (1380g)
WR-17	Reference 1	19	Composite 14 (19 crab)	6 (913g)
WR-18	Reference 1	14	Composite 15 (14 crab)	6 (408g)
WR-19	Reference 1	15	Composite 16 (15 crab)	12 (1442g)
WR-21/22/24	Reference 4	8	Composite 17 (8 crab)	4 (350g)
WR-25	Reference 3	5	Composite 18 (5 crab)	5 (40)
WR-26	Reference 3	22	Composite 19 (22 crab)	6 (258g)
WR-27	Reference 3	10	Composite 20 (10 crab)	10 (1193g)
<b>Total</b>	<b>Reference</b>	<b>123</b>		<b>73 (6096)</b>

Note: - Numbers of crab for taste tests is approximate because crab legs were often broken off carapace

## 6.2.2 Body Burden

Samples were delivered frozen to Maxxam Analytics in Halifax, Nova Scotia, and processed for the analytes listed in Table 6-4. Analytical methods and QA/QC procedures for these tests are provided in Appendix C-2.

**Table 6-4 Body Burden Variables (2000 to 2005)**

Variables	Method	2000 EQL	2002 EQL	2004 EQL	2005 EQL	Units
<b>Hydrocarbons</b>						
>C <sub>10</sub> -C <sub>21</sub>	GC/FID	15	15	15	15	mg/kg
>C <sub>21</sub> -C <sub>32</sub>	GC/FID	15	15	15	15	mg/kg
<b>PAHs</b>						
1-Chloronaphthalene	GC/MS	NA	NA	0.05	0.05	mg/kg
2-Chloronaphthalene	GC/MS	NA	NA	0.05	0.05	mg/kg
1-Methylnaphthalene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
2-Methylnaphthalene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Acenaphthene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Acenaphthylene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Anthracene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Benzo[a]anthracene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Benzo[a]pyrene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Benzo[b]fluoranthene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Benzo[ghi]perylene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Benzo[k]fluoranthene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Chrysene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Dibenz[a,h]anthracene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Fluoranthene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Fluorene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Indeno[1,2,3-cd]pyrene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Naphthalene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Perylene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Phenanthrene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
Pyrene	GC/MS	0.05	0.05	0.05	0.05	mg/kg
<b>Metals</b>						
Aluminum	ICP-MS	2.5	2.5	2.5	2.5	mg/kg
Antimony	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Arsenic	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Barium	ICP-MS	1.5	1.5	1.5	1.5	mg/kg
Beryllium	ICP-MS	1.5	1.5	0.5	0.5	mg/kg

Variables	Method	2000 EQL	2002 EQL	2004 EQL	2005 EQL	Units
Boron	ICP-MS	1.5	1.5	1.5	1.5	mg/kg
Cadmium	GFAAS	0.08	0.05	0.05	0.05	mg/kg
Chromium	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Cobalt	ICP-MS	0.2	0.2	0.2	0.2	mg/kg
Copper	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Iron	ICP-MS	5	5	15	15	mg/kg
Lead	ICP-MS	0.18	0.18	0.18	0.18	mg/kg
Lithium	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Manganese	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Mercury	CVAA	0.01	0.01	0.01	0.01	mg/kg
Molybdenum	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Nickel	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Selenium	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Silver	ICP-MS	0.12	0.12	0.12	0.12	mg/kg
Strontium	ICP-MS	1.5	1.5	1.5	1.5	mg/kg
Thallium	ICP-MS	0.02	0.02	0.02	0.02	mg/kg
Tin	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Uranium	ICP-MS	0.02	0.02	0.02	0.02	mg/kg
Vanadium	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
Zinc	ICP-MS	0.5	0.5	0.5	0.5	mg/kg
<b>Other</b>						
Percent Lipids/Crude Fat	PEI FTC/ AOAC922.06	0.1	0.5	0.5	0.5	%
Moisture	Grav.	0.1	0.1	0.1	0.1	%

- Notes:
- The EQL is the lowest concentration that can be reliably detected within specified limits of precision and accuracy during routine laboratory operating conditions. EQLs may vary from year to year because of methods improvement and because instruments are checked for precision and accuracy every year as part of QA/QC procedures.
  - NA = Not Analyzed

### 6.2.3 Taste Tests

Plaice and crab samples were delivered frozen to the Fisheries and Marine Institute of Memorial University of Newfoundland for sensory evaluation, using taste panels and triangle and hedonic scaling test procedures. Since no procedures have been established to compare multiple Reference Areas to one Study Area, samples were randomly selected from each of the four Reference Areas to generate one set of Reference Area samples to be compared to Study Area samples.

Frozen plaice samples were thawed for 24 hours at 2°C, removed from plastic bags and homogenized in a food processor. Samples were allocated to either the triangle taste test or the hedonic scaling test. Samples were enclosed in individual aluminum foil packets (Figure 6-1), labeled with a predetermined random three-digit code and cooked in a convection oven at 82°C for 11 minutes. Plaice samples were served at approximately 35°C.



**Figure 6-1 Plaice Taste Test Preparations**

Frozen crab samples were cooked, shucked of meat and stored overnight at 4°C in a plastic container. All meat was homogenized in a food processor and allocated to either the triangle taste test or the hedonic scaling test. Crab was served to taste panelists in plastic cups at room temperature.

Each panel included 24 untrained panelists who were provided with score sheets (Figures 6-2 and 6-3) and briefed on the presentation of samples prior to taste tests. Panelists were instructed that samples were being tested for uncharacteristic odour or taste and that grit, cartilage and texture should not be considered in their assessment. Panelists were also instructed not to communicate with each and to leave immediately upon completion of the taste tests.



**QUESTIONNAIRE FOR TRIANGLE TEST**

Name: \_\_\_\_\_ Date/Time: \_\_\_\_\_

Product: American Plaice

Two the samples in the order indicated and identify the odd sample.

1. Taste the samples in the order indicated and identify the odd sample.  
You must choose one of the samples.

Code	Check Odd Sample
214 _____	
594 _____	
733 _____	

2. Comments:

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

**Figure 6-2 Questionnaire for Sensory Evaluation by Triangle Test**

**QUESTIONNAIRE FOR HEDONIC SCALING**

Name: \_\_\_\_\_ Date/Time: \_\_\_\_\_

Product: American Plaice

1. Taste these samples and check how much you like of dislike each one.

<p><u>619</u></p> <p><input type="checkbox"/> Like extremely</p> <p><input type="checkbox"/> like very much</p> <p><input type="checkbox"/> like moderately</p> <p><input type="checkbox"/> like slightly</p> <p><input type="checkbox"/> neither like or dislike</p> <p><input type="checkbox"/> dislike slightly</p> <p><input type="checkbox"/> dislike moderately</p> <p><input type="checkbox"/> dislike very much</p> <p><input type="checkbox"/> dislike extremely</p>	<p><u>835</u></p> <p><input type="checkbox"/> Like extremely</p> <p><input type="checkbox"/> like very much</p> <p><input type="checkbox"/> like moderately</p> <p><input type="checkbox"/> like slightly</p> <p><input type="checkbox"/> neither like or dislike</p> <p><input type="checkbox"/> dislike slightly</p> <p><input type="checkbox"/> dislike moderately</p> <p><input type="checkbox"/> dislike very much</p> <p><input type="checkbox"/> dislike extremely</p>
---	---

2. Comments:

\_\_\_\_\_

\_\_\_\_\_

\_\_\_\_\_

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\_\_\_\_\_

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\_\_\_\_\_

**Figure 6-3 Questionnaire for Sensory Evaluation by Hedonic Scaling**

For the triangle test, panelists were presented with a three-sample set (triangle) of samples and asked to identify the sample that was different from the others. Half of the panelists received sets composed of two samples from Treatment A (Study Area) and one from Treatment B (Reference Areas). The other panelists received sets composed of one sample from Treatment A and two from Treatment B. There were six possible orders in which the samples were presented to panelists, after Botta (1994): ABB, AAB, ABA, BAA, BBA, and BAB.

The rest of the samples were used for hedonic scaling tests. In this test, one sample from the Study Area and one from the Reference Areas were presented to panelists. Panelists were instructed to rate how much they liked or disliked each sample on the form provided to them. A nine-point hedonic scale was used, with ratings ranging from “like extremely” (9) to “dislike extremely” (1) (see Figure 6-3 for full range of ratings).

## 6.2.4 Fish Health Indicators

### 6.2.4.1 Mixed Function Oxygenase

MFO induction was assessed in liver samples of plaice as 7-ethoxyresorufin O-deethylase (EROD) activity according to the method of Pohl and Fouts (1980) as modified by Porter et al. (1989).

#### Sample preparation

Liver samples were thawed on ice within four weeks of storage at  $-65^{\circ}\text{C}$  and homogenized in four volumes of 50 mM Tris buffer, pH 7.5, (1 gram liver to 4 ml buffer) using at least ten passes of a glass Ten Broek hand homogenizer. Homogenates were centrifuged at 9,000 g for 15 minutes at  $4^{\circ}\text{C}$  and the post-mitochondrial supernatant (S9 fraction) frozen in triplicate at  $-65^{\circ}\text{C}$  until assayed.

All liver samples were held and processed under the same storage and assay conditions. Assays were carried out within four weeks of storage of S9 fractions.

#### EROD assay

The reaction mixture, final volume of 1 ml, contained 50 mM Tris buffer, pH 7.5, 2  $\mu\text{M}$  ethoxyresorufin (Sigma) dissolved in dimethyl sulphoxide, 0.15 mM NADPH and 20  $\mu\text{l}$  of S9 protein (diluted five times). After a 15-minute incubation at  $27^{\circ}\text{C}$ , the reaction was stopped with 2 ml of methanol (HPLC grade) and samples were centrifuged (3,600 g for five minutes) in order to remove the protein precipitate. The fluorescence of resorufin formed in the supernatant was measured at an excitation wavelength of 550 nm and an emission wavelength of 580 nm using a Perkin-Elmer LS-5 fluorescence spectrophotometer. Blanks were performed as above with methanol added before the incubation. All the samples were run in duplicate. Protein concentration was determined using the Lowry protein method (Lowry et al. 1951) with bovine serum albumin as standard. The rate of enzyme activity in pmol/min/mg protein was obtained from the regression of fluorescence against standard concentrations of resorufin. One low and one high resorufin standard were prepared daily from a stock solution and run with each batch of samples to check the standard curve.

### 6.2.4.2 Haematology

Blood smears were stained with Giemsa stain and examined with a Wild Leitz Aristoplan bright field microscope for identifying different types of cells based on their general form and affinity to the dye (Ellis 1976).

Size, shape and degree of haemoglobinization of red blood cells were examined and recorded.

Differential blood cell counts were performed on lymphocytes, neutrophils and thrombocytes and expressed as a percentage of each type of cells on 200 white blood cells counted. Cells were counted under 400x magnification (or oil immersion, 1000x, when necessary) in fields along a row commencing from the front edge of the smear continuing parallel to the slide edge until the total number of cells were counted.

### 6.2.4.3 Histopathology

Fixed liver and gill samples were processed by standard histological methods (Lynch et al. 1969) using a Tissue-Tek® VIP Processor. A graded ethyl alcohol series of 70%, 80%, 95%, and two changes of 100%, were used for dehydration of the samples. The livers were then cleared in three changes of chloroform. Finally, the tissues were impregnated with three changes of molten embedding media, Tissue Prep 2™. The processed tissues were embedded in steel molds using molten embedding media, and topped with labeled embedding rings. After cooling, the hardened blocks of embedded tissues were removed from their base molds. The blocks were then trimmed of excess wax. Sections were cut at 6 µm on a Leitz microtome, floated on a 47°C water bath containing gelatin, and then picked up on labeled microscope slides. After air drying, slides were fixed at 60°C for approximately two hours to remove most of the embedding media and allow the tissue to adhere properly to the slide. Sections were stained using Mayers Haematoxylin and Eosin method (Luna 1968). Coverslips were applied using Entellan® and the slides were left to air dry and harden overnight.

Histological examination of each tissue was conducted by the same investigator. One slide with four to six sections was examined per fish. If an abnormality was found in a section, the other sections were checked for the same abnormality. To minimize interpretive bias, a “blind” system in which the examiner is not aware of the site of capture of specimen was used. This is accomplished by using a “pathology” number on the slide label generated from a random number table matched with the actual specimen number.

#### Liver

All liver samples were assessed microscopically for the presence of different lesions previously identified as having a putative chemical aetiology in fish (e.g., Myers et al. 1987; Myers and Fournie 2002). Among them were:

- |                          |                             |
|--------------------------|-----------------------------|
| 1. Nuclear pleomorphism  | 6. Hepatocellular carcinoma |
| 2. Megalocytic hepatitis | 7. Cholangioma              |
| 3. Eosinophilic foci     | 8. Cholangiofibrosis        |
| 4. Basophilic foci       | 9. Macrophage aggregates    |
| 5. Clear cell foci       | 10. Hydropic vacuolation    |

Any other observations were also recorded. Among them, hepatocellular vacuolation, parasitic infestation of the biliary system and inflammatory response.

Lesions (except macrophage aggregates and inflammatory response) were recorded for each fish as not detected (0) or detected (1).

Macrophage aggregation was recorded on a relative scale from 0 to 7 and prevalence was calculated for fish showing a moderate to high aggregation (3 or higher on the scale). Inflammatory response was recorded on a relative scale from 0 to 3 (0-absent, 1-mild, 2-moderate and 3-heavy).

The percentage of fish affected by each type of lesions or prevalence of lesion was then calculated.

**Gill**

Each gill sample was examined microscopically, first under low power (x63) for a general overview of the entire section and to record any abnormalities or parasites present. Next, five randomly selected fields were read at x250 magnification for the presence of established gill lesions (Mallat 1985).

For each field, the total number of secondary lamellae were counted and recorded. Each lamella was then examined quantitatively for six different stages (Table 6-5).

**Table 6-5 Stages for Gill Lamella**

Stage 1 - Thin lamellae	Operationally defined here as secondary lamellae having a one-cell thick epithelial layer, with the base between two secondary lamellae having a three to five-cell thick epithelial layer.
Stage 2 - Distal hyperplasia	Thickening of the epithelium from the basal end and running almost the entire length of secondary lamellae (which may also appear misshapen).
Stage 3 - Epithelial lifting	Separation of the epithelial layer from the basement membrane.
Stage 4 - Clubbing	Swelling of the distal end of secondary lamellae which occurs in two different forms: a) <i>tip hyperplasia</i> - thickening of the epithelium at the very tip of lamellae giving the appearance of a club; and b) <i>telangiectasis</i> - a swelling without rupture of the capillary at the distal end of lamellae (i.e., aneurism).
Stage 5 - Basal hyperplasia	Thickening of the epithelium near the base of secondary lamellae where they meet the primary filament.
Stage 6 - Fusion	Fusion of two or more lamellae.

Note: - Stages do not follow in any specific order

Results for each fish were expressed as the percentage of lamellae presenting the stage in relation to the total number of lamellae counted in the fields.

The degree of oedema present, if any, was recorded on a 0 to 3 relative scale (0-absent, 1-light, 2-moderate and 3-heavy).

**6.3 Data Analysis**

For most analyses except taste tests, the Commercial Fish component of the 2005 White Rose EEM program used a multiple-reference design with three or four Reference Areas and two sub-Areas, North and South, within the Study Area. Three comparisons were of interest:

- Study versus Reference Areas (SR)
- Between Study Areas (BS)
- Among Reference Areas (AR)

The modified nested Analysis of Variance (ANOVA) model in Table 6-6 was used for analysis of continuous variables. The SR and BS contrasts are tested against the variance (MS) among Reference Areas or MS(AR), which is a measure of natural variance among Areas. The SR contrast compares the mean for the Northern and Southern portion of the Study Area to the mean for the Reference Areas, and provides a test for overall project effects. The BS contrast compares the difference between the Northern and Southern portion of the Study Area to variances among the Reference

Areas. The Study Area was split into north and south parts to provide even coverage, and not to test for smaller-scale natural or project-related differences within the overall Study Area. However, the BS contrast can provide additional information, and in some cases, splitting the Study Area can increase the power of tests of the SR contrast. The AR contrast is tested against the variance among replicates (composites for most analyses) within Areas (MSE). This test is equivalent to an ANOVA comparing the Reference Areas with composites as replicates within Areas, except that variance among replicates within the Northern and Southern portion of the Study Area is incorporated into the MSE.

**Table 6-6 Modified Nested ANOVA Model for Analysis of Multiple-Reference Design**

Source/Term	df	Mean Square (MS)	F
<b>Among Areas</b>			
Study versus Reference (SR)	1	MS(SR)	MS(SR)/MS(AR)
Between Study (BS)	1	MS(BS)	MS(BS)/MS(AR)
Among References (AR)	$r-1$	MS(AR)	MS(AR)/MSE
<b>Within Areas</b>			
Among composites	$N-5$	MSE	

Notes: - df = degrees of freedom  
 -  $r$  = number of Reference Areas ( $r = 3$  for analyses of crab and  $r = 4$  for analyses of plaice)  
 -  $N$  = total number of composites

The model in Table 6-6 is referred to as a “modified” nested ANOVA because it is unconventional, with no replicate “Areas” within the Northern and Southern portion of the Study Area. There are reasonable alternative models and significance tests (see Quinn and Keough (2002) for an extended discussion). With three replicate Reference Areas for crab and four for plaice, tests of the SR and BS contrasts will not be powerful. If the added natural variance among Reference Areas is “small” (i.e., MS(AR) is less than or similar to MSE), power can be increased by testing the SR and BS contrasts against the MSE, which is based on approximately 20 composites. Quinn and Keough (2002) discuss the issue of when to pool higher- and lower-order terms, or test against lower-order terms (e.g., MSE), in nested and other complex ANOVA. They recommend testing against lower-order terms when  $p \geq 0.25$  for higher-order terms such as MS(AR). Their recommendation was adopted in this report for interpretation of results. However, other authors have recommended using  $p$  from 0.05 to  $\geq 0.50$  to define when higher-order terms are “small”, so  $p$  for tests of the SR and BS contrasts against both MS(AR) and MSE are provided in this report.

**6.3.1 Biological Characteristics**

Biological Characteristics (morphometric and life history characteristics) of crab and plaice were analyzed primarily to determine if there were differences among composites that could affect results of body burden analyses. The analyses of Biological Characteristics also provided basic biological information on the two species.

**6.3.1.1 Crab**

Biological Characteristics of crab included carapace width and claw height (i.e., size), and frequency of recent moult based on the shell condition index (see Appendix C-1). Recent moults included crab with shell condition index values of 1 or 2. Non-recent moults included crab with condition index values of 6 (probably one year since moult),

and 3 or 4 (two or more years since moult). Values other than 1 to 4 and 6 were not observed.

The first step was to determine if there was added variance among composites within Areas. Variance among composites is small-scale spatial variance among trawl locations. The nested ANOVA in Table 6-6, with a third level added (variance among individual crab within composites), was used for the analysis. The variance among composites within Areas is tested against the variance among crab within composites. For individual crab, a score of 0 was assigned to recent moults and a score of 1 was assigned to non-recent moults.

For all three biological variables, there was significant ( $p < 0.0001$ ) added variance among composites within Areas. Therefore, mean carapace width, claw height and frequencies of recent moults were calculated for each composite, and the composite values analyzed in the nested ANOVA in Table 6-6.

Spearman rank correlations ( $r_s$ ) were calculated among the three biological variables based on individual and composite values. Correlations were calculated over all Areas pooled, and then separately for the pooled Reference Areas and the pooled Study Areas.

The statistical analyses described above excluded Reference Area 4 because there was only one composite of eight (8) crab from that Area. However, Reference Area 4 values were included in plots and summary tables for comparison with values from other Reference Areas.

Analyses of Biological Characteristics were restricted to crab used for body burden analyses in 2005. Formal comparisons between 2004 and 2005 were not conducted, but some differences between results for the two years were briefly noted.

### 6.3.1.2 Plaice

In this section, analyses of plaice Biological Characteristics were restricted to gutted weight (i.e., size). Immature and mature females and males were pooled for the analyses, since they were pooled within composites. The primary objective was to determine if there were size differences that might affect results of body burden analyses. Appendix C-3 provides more extensive analyses of a larger suite of biological variables (length, age, body weight, liver and gonad weight) for plaice.

Analyses were conducted on composite mean weights. Distributions of individual weights within composites were rarely normal. Instead, they were usually bimodal, since immature fish were smaller than mature fish and males smaller than females. Distributions of individual weights were also truncated at the left (low) end because fish smaller than 250 mm in length were released and not retained for body burden and health analyses. Composite mean weights were compared among Areas using the nested ANOVA in Table 6-6. Differences between 2005 and 2004 results were briefly noted.

## 6.3.2 Body Burden

### 6.3.2.1 Crab

#### Analyses of 2005 Data

Body burden variables analyzed were moisture content, fat (lipid) content and dry weight concentrations of eight frequently detected metals (arsenic, boron, copper, mercury, selenium, silver, strontium and zinc). Fat content was not measured on one composite from Reference Area 2 and one composite from Reference Area 3 because of insufficient sample volume. Variable values less than EQL were set at EQL rather than  $\frac{1}{2}$  EQL. For the sediment component of this report and for analyses of body burdens in the 2004 report (Husky Energy 2005), values less than EQL were set at  $\frac{1}{2}$  EQL. However, for body burdens, the two-fold difference between EQL versus  $\frac{1}{2}$  EQL was larger than most differences in detectable concentrations within and among Areas, and using  $\frac{1}{2}$  EQL to replace values less than EQL could potentially bias analyses and results.

A summary measure of metal concentrations was derived using (PCA<sup>7</sup>). Metal concentrations were log<sub>10</sub> transformed prior to conducting the PCA. The PCA included all samples from both 2004 and 2005, since PC1 scores were compared between years (see below).

Fat content, moisture content, Metals PC1 scores and untransformed concentrations of the eight metals frequently detected in 2005 were analyzed in the nested ANOVA in Table 6-6. Fat content and Metals PC1 scores were rank-transformed to remove the effects of outliers. Rank correlations were also calculated among body burden variables and between them and the three biological variables (carapace width, claw height and % recent moult).

#### Comparison of 2004 Versus 2005 Results

Body burden results from 2004 and 2005 were compared in the RM ANOVA in Table 6-7, which can be considered the RM or multi-year version of the nested ANOVA in Table 6-6. The Study Area was treated as a single Area because it was not distinctly split into North and South Areas in 2004 (see Figures 1-7 and 1-8, Section 1). Values analyzed were Area means to simplify analysis and interpretation. The ANOVA has limited power with only two years and four Areas, but sample sizes and power will rapidly increase as more years are added in future EEM programs.

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<sup>7</sup> PCA identifies the major axis of covariance (Principal Component or PC1) among the original variables (concentrations of the eight metals). PC1 is also the major axis of variance among samples (i.e., composites). PCA then identifies lesser (minor) axes of variance, each perpendicular to, and uncorrelated with, PC1 and each other. PC2 will account for more variance than PC3, PC3 will account for more variance than PC4, and so on. Positions of samples along any axis or PC can be defined by scores, which are weighted means or sums of the original variables. The scores are scaled so that the mean is 0 and the variance and standard deviation (SD) are 1. The scores can be used as summary variable values for further analyses.



**Table 6-7 Repeated Measures (RM) Used for Comparison of Body Burden Variables Between 2004 and 2005**

Source/Term	df	F
<i>Between Areas</i>		
Study versus Reference (SR)	1	MS(SR)/MS(AR)
Among References (AR)	$r-1$	MS(AR)/MS(Year × AR)
<i>Within Areas</i>		
Year (Y)	1	MS(Y)/MS(Year × AR)
Year × SR	1	MS(Y × SR)/MS(Year × AR)
Year × AR	$r-1$	Not tested

Notes: - df = degrees of freedom  
 - MS = Mean Square  
 -  $r$  = number of Reference Areas

The Between Areas terms in the RM ANOVA compare averages over the two years among Areas. These are tests for persistent differences among Areas over time. The Study versus Reference (SR) difference is tested against the variance (MS) among Reference Areas, as in the nested ANOVA used for analysis of 2005 data (Table 6-6). The differences among Reference Areas represent carry-over effects.

The Within Areas terms in the RM ANOVA test for consistent differences between years in all Areas (Year term), and changes in the SR difference between years (Year x SR term). The error term for the Within Areas tests, and the Between Areas AR contrast, is the Year x AR interaction (changes in differences among Reference Areas between years).

Body burden variables compared between years were moisture content, fat content, Metals PC1 and concentrations of the eight metals analyzed for 2005. Analyses were conducted with and without Reference Area 4, which was represented by a single composite in 2005. Values less than EQL were set at EQL.

### 6.3.2.2 Plaice

#### Analyses of 2005 Data

Body burden data from composite samples were available for both liver and fillet tissue. Variables analyzed for liver were moisture and fat content, concentrations of eight metals detected in every composite (arsenic, cadmium, copper, iron, manganese, mercury, selenium, zinc), Metals PC1 derived from log-transformed concentrations of the eight metals, and  $>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HC concentrations.

Fat content could not be measured on one liver composite from Reference Area 2 and three composites from the Northern portion of the Study Area because of insufficient sample volume.

Variables analyzed for fillets were moisture and fat content, and concentrations of arsenic, mercury and zinc (detected in every composite).

Body burden variables for liver and fillets were compared among Areas in the nested ANOVA in Table 6-6. Liver  $>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HC concentrations were rank-transformed to remove the effects of outliers.

### **Comparison of 2005 versus 2004 Results**

Plaise body burden results from 2004 and 2005 were compared in the RM ANOVA in Table 6-7. Variables analyzed were the same as those analyzed for 2005.

#### **6.3.3 Taste Tests**

Unlike analyses on Biological Characteristics (Section 6.3.1), body burdens (Section 6.3.2) and health (Section 6.3.4), triangle tests and hedonic scaling tests compared Study Area samples to pooled Reference Area samples (see Section 6.2.3).

The triangle test datum is the number of correct sample identifications over the number of panelists. This value was calculated and compared to values in Appendix C-4 (after Larmond 1977) to determine statistical significance. For a panel size of 24, a statistically significant discrimination between Areas (at  $\alpha = 0.05$ ) would require that 13 panelists correctly identify samples.

Hedonic scaling results were processed in ANOVA and presented graphically in a frequency histogram.

Ancillary comments from panelists were tabulated and assessed for both tests.

#### **6.3.4 Fish Health Indicators**

For fish health, a multiple-reference design with four Reference Areas and a North and South Study Area was used in analyses and three comparisons, Among Reference Areas, between the Northern and Southern portion of the Study Area and between the Study Area and Reference Areas were conducted similar to comparisons detailed in Sections 6.3.1 and 6.3.2. Details on these statistical methods are provided in Appendix C-3 (Annex B).

### **6.4 Results**

#### **6.4.1 Biological Characteristics**

##### **6.4.1.1 Crab**

Shell condition index values for the 272 crab used for body burden analyses in 2005 are provided in Table 6-8. Most index values were 2 (recent moult) or 6 (moulted last year), with frequencies of these two values approximately equal. Frequencies of recent moults for the Northern and Southern portion of the Study Area and all Reference Areas pooled were similar and approximately 50%. Frequencies of recent moults were highest in Reference Area 3 and lowest in Reference Area 2.

**Table 6-8 Frequencies of Crab Shell Condition Index Values (2005)**

Moult year	Index value	Area								Total
		Ref 1	Ref 2	Ref 3	Ref 4	All Refs	North Study	South Study	Both Study	
Recent (0)	1	0	0	0	0	0	0	1	1	1
	2	20	7	28	5	60	40	40	80	140
Total (No.)		20	7	28	5	60	40	41	81	141
(%)		53	23	76	63	53	49	53	51	52
Not recent (-1+)	6	18	23	7	3	51	41	35	76	127
Last year (-1)	3	0	0	2	0	2	0	1	1	3
Previous (-2+)	4	0	0	0	0	0	1	0	1	1
Total (No.)		18	23	9	3	53	42	36	78	131
(%)		47	77	24	38	47	51	47	49	48
Grand total (No.)		38	30	37	8	113	82	77	159	272

Notes: - Moult years: 0 = 2005; -1 = 2004; -2+ = 2003 or earlier  
 - Values are numbers of crab unless otherwise indicated

Summary statistics for composite means are provided in Table 6-9. Mean and median carapace widths were approximately 70 mm in 2005 versus approximately 100 mm in 2004. In 2005, crab from the Northern portion of the Study Area and Reference Area 1 were larger than crab in other Areas. Area differences in claw height were similar to those for carapace width, since the two size measures were correlated (see below). CVs for the two size measures were greater (i.e., size was more variable) in Reference Areas 2 and 3 than in other Areas.

**Table 6-9 Summary Statistics for Biological Characteristics of Crab Based on Composite Means (2005)**

Variable	Area	n	Min	Max	Median	Mean	SD	CV (%)
Carapace width (mm)	Reference Area 1	3	62	91	89	81	16	20
	Reference Area 2	3	50	94	57	67	23	35
	Reference Area 3	3	51	94	54	67	24	36
	Reference means					72		
	Reference Area 4	1				70		
	North Study Area	5	66	104	82	83	16	20
	South Study Area	5	55	82	64	64	11	17
	Study means					73		
Claw height (mm)	Reference Area 1	3	12.1	20.2	19.4	17.2	4.5	26
	Reference Area 2	3	8.1	21.4	9.8	13.1	7.2	55
	Reference Area 3	3	8.4	21.5	9.3	13.1	7.3	56
	Reference means					14.5		
	Reference Area 4	1				15.7		
	North Study Area	5	13.3	24.2	17.6	18.0	4.6	25
	South Study Area	5	9.4	18.4	11.1	12.2	3.6	30
	Study means					15.1		
% recent moult	Reference Area 1	3	33	71	47	50	19	
	Reference Area 2	3	0	50	27	26	25	
	Reference Area 3	3	70	80	77	76	5	
	Reference means					51		
	Reference Area 4	1				63		
	North Study Area	5	0	100	46	49	50	
	South Study Area	5	7	95	44	52	33	
	Study means					51		

Notes: - CV = Coefficient of Variation (SD as % of mean)  
 - Reference Area 4 means are not included in overall Reference means because they were not included in ANOVA comparisons among Reference Areas and between the Study Area and Reference Areas

Differences in frequencies of recent moults based on composite means in Table 6-9 were similar to those for individual crab in Table 6-8. The frequencies within Areas in the two tables do not match exactly because values based on individuals weight each composite by sample size whereas values based on composite means weight each composite equally. CVs are not provided for % recent moult because composite means could be expressed as either % recent moult or % non-recent moult (100-% recent moult; SDs remain the same). SDs were greater in the Northern portion of the Study Area and lower in Reference Area 3 than in other Areas. The differences in variance among Areas may have affected analyses in ANOVA but could not be removed by transformations.

Table 6-9 also includes values from the single Reference Area 4 composite of eight (8) crab. The Reference Area 4 crab were intermediate in size in 2005, whereas Reference Area 4 crab in 2004 were larger than crab from other Areas (Husky Energy 2005).

None of the differences in Biological Characteristics among Areas noted above were statistically significant when tested in the modified nested ANOVA (Table 6-10). All *p* values for the Study versus Reference contrast were much greater than 0.05. The lowest *p* values (all > 0.10) were obtained for comparisons of size (carapace width and claw height) between the Northern and Southern portion of the Study Area.

**Table 6-10 Results of Modified Nested ANOVA Comparing Crab Biological Characteristics Among Areas (2005)**

Variable	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
Carapace width	0.547	0.173	<i>0.120</i>	0.799	<i>0.821</i>
Claw height	0.551	0.159	<i>0.105</i>	0.764	<i>0.791</i>
% recent moult	0.236	<i>0.922</i>	0.890	<i>1.000</i>	0.992

Notes: - Reference Area 4 (1 composite) excluded  
 - MSE = variance among composites within Areas; MS(AR) = variance among Reference Areas  
 - Italics indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

As expected, the two size variables (carapace width and claw height) were significantly and strongly positively correlated among individual crab and among composite means (Table 6-11). For individual crab, size and % recent moult were negatively correlated for all Areas combined and for the Reference Areas combined, indicating that smaller Reference Area crab were more likely to have moulted in 2005. In the Study Area(s), size and % recent moult were uncorrelated. Overall and Reference Area correlations between size and % recent moult for composite means were weaker and not significant. These results indicate that size and frequencies of recent moult were correlated within rather than among composites, specifically within Reference Area composites. In 2004, size and % recent moult were also negatively correlated, but correlations among composite means were stronger than correlations among individuals, and correlations were similar for the Reference and Study Areas (Husky Energy 2005).

**Table 6-11 Spearman Rank Correlations ( $r_s$ ) Among Crab Biological Variables (2005)**

Values	Areas	Carapace width-claw height		Carapace width-% recent moult		Claw height-% recent moult	
		<i>n</i>	<i>r<sub>s</sub></i>	<i>n</i>	<i>r<sub>s</sub></i>	<i>n</i>	<i>r<sub>s</sub></i>
Individual crab	All	246	<b>0.945**</b>	264	<b>-0.300**</b>	246	<b>-0.298**</b>
	Reference	96	<b>0.953**</b>	105	<b>-0.639**</b>	96	<b>-0.636**</b>
	Study	150	<b>0.935**</b>	159	-0.050	150	-0.065
Composite means	All	19	<b>0.995**</b>	19	0.017	19	0.026
	Reference	9	<b>0.983**</b>	9	-0.200	9	-0.183
	Study	10	<b>1.000**</b>	10	0.238	10	0.238

Notes: - Reference Area 4 (1 composite of 8 crab) excluded  
 - \*  $p \leq 0.05$ ; \*\*  $p \leq 0.01$ ;  $r_s$  at  $p \leq 0.05$  in bold

**6.4.1.2 Plaice**

Summary statistics for composite mean gutted weights of plaice are provided in Table 6-12. Females accounted for 86% of the 180 plaice used for body burden and health analyses. Approximately 65% of the females were mature. Therefore, most composites were a mix of larger mature females (and the occasional large male) that could exceed 1,000 g, and smaller males and immature females that were usually less than 300 g. Reference Area 3 fish and composites were the exception. All fish caught there were large mature females and mean weight was much greater than for other Areas (Table 6-12). Consequently, the Among References contrast in the modified nested ANOVA was highly significant and the Study versus Reference and Between Study contrasts were not significant when tested against the large variance among Reference Areas (Table 6-13). The largest size difference among the five Areas other than Reference Area 3 was between the Northern and Southern portion of the Study Area, with fish larger in the South (Table 6-12). The large size difference between Reference Area 3 fish and fish from other Areas may have affected some of the body burden results (Section 6.4.2.2), and was much greater than any size difference observed in 2004 (Husky Energy 2005).

**Table 6-12 Summary Statistics for Plaice Gutted Weight, Based on Composite Means (2005)**

Area	<i>n</i>	Min	Max	Median	Mean	SD	CV (%)
Reference Area 1	3	361	495	439	431	67	16
Reference 2	3	240	507	383	377	134	36
Reference 3	3	840	1,029	918	929	95	10
Reference 4	3	383	494	448	442	56	13
Reference means					545		
North Study	5	193	514	456	394	133	34
South Study	5	479	796	584	603	122	20
Study means					498		

**Table 6-13 Results of Modified Nested ANOVA Comparing Plaice Composite Mean Guttled Weights Among Areas (2005)**

<i>p</i> values				
Among References	Between Study		Study versus Reference	
Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
<0.001	<i>0.513</i>	0.009	<i>0.824</i>	0.345

Notes: - MSE = variance among composites within Areas; MS(AR) = variance among Reference Areas  
 - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

Results were similar for biological variables measured on mature and immature females tested in conjunction with fish health analysis (Appendix C-3). Size, age and condition of mature females did not differ significantly between the Study versus Reference Areas. Mature females from Reference Area 3 were significantly larger (and older) than mature females from other Areas. Size and condition factor (weight relative to length) of immature females were significantly greater in the Southern portion versus the Northern portion of the Study Area, but the overall difference between the Study versus Reference Areas was not significant. Reference Area 4 immature females were larger than immature females from Reference Areas 1 and 2; no immature females were collected from Reference Area 3.

**6.4.2 Body Burden**

**6.4.2.1 Crab**

Summary statistics for concentrations of detected substances in crab claw composites in 2004 and 2005 are provided in Table 6-14. Raw data for 2005 are provided in Appendix C-2.

**Table 6-14 Summary Statistics for Crab Body Burden (2004, 2005)**

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
Arsenic	2004	Reference Area 1	3	0	6.90	7.80	7.50	7.40	0.46	6
		Reference Area 2	3	0	6.80	10.00	9.60	8.80	1.74	20
		Reference Area 3	2	0	8.50	8.60	8.55	8.55	0.07	1
		Reference Area 4	3	0	11.00	13.00	11.00	11.70	1.15	10
		Reference Means					9.16	9.10		
	Study Area	10	0	4.80	12.00	8.55	8.71	2.44	28	
	2005	Reference Area 1	3	0	6.11	9.46	7.84	7.80	1.68	21
		Reference Area 2	3	0	5.22	8.51	7.55	7.09	1.69	22
		Reference Area 3	3	0	7.38	8.38	7.93	7.90	0.50	6
		Reference Area 4	1	0	9.02	9.02	9.02	9.02		
		Reference Means					8.09	7.95		
		Study Area North	5	0	6.46	7.64	6.93	7.04	0.50	7
		Study Area South	5	0	5.59	6.81	6.16	6.26	0.48	8
Study Means						6.55	6.65			
Boron	2004	Reference Area 1	3	0	1.90	2.50	2.30	2.23	0.31	14
		Reference Area 2	3	1	<1.5	2.80	1.90			
		Reference Area 3	2	0	1.70	2.30	2.00	2.00	0.42	21
		Reference Area 4	3	1	<1.5	2.00	1.90			
		Reference Means					2.03			
	Study Area	10	1	<1.5	3.20	1.90				
	2005	Reference Area 1	3	1	<1.5	2.40	1.70			0
		Reference Area 2	3	0	2.10	4.70	3.30	3.37	1.30	39

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
Boron	2005	Reference Area 3	3	0	2.30	4.20	3.80	3.43	1.00	26
		Reference Area 4	1	0	3.30	3.30	3.30	3.30	.	
		Reference Means					3.03			
		Study Area North	5	0	2.40	4.10	3.20	3.22	0.64	20
		Study Area South	5	1	<1.5	3.50	2.30			
		Study Means					2.75			
Cadmium	2004	Reference Area 1	3	2	<0.05	0.070	<0.05			
		Reference Area 2	3	2	<0.05	0.050	<0.05			
		Reference Area 3	2	1	<0.05	0.050	<0.05			
		Reference Area 4	3	1	<0.05	0.100	0.050			
		Reference Means								
		Study Area	10	3	<0.05	0.100	0.050			
	2005	Reference Area 1	3	3	<0.05	<0.05	<0.05			
		Reference Area 2	3	3	<0.05	<0.05	<0.05			
		Reference Area 3	3	3	<0.05	<0.05	<0.05			
		Reference Area 4	1	1	0.090	0.090	0.090	0.090		
		Reference Means								
		Study Area North	5	5	<0.05	<0.05	<0.05			
		Study Area South	5	3	<0.05	0.054	<0.05			
		Study Means								
Copper	2004	Reference Area 1	3	0	2.90	4.00	3.20	3.37	0.57	17
		Reference Area 2	3	0	3.10	5.80	5.30	4.73	1.44	30
		Reference Area 3	2	0	3.20	3.80	3.50	3.50	0.42	12
		Reference Area 4	3	0	4.20	5.10	4.70	4.67	0.45	10
		Reference Means					4.18	4.07		
		Study Area	10	0	2.90	4.80	3.90	3.94	0.63	16
	2005	Reference Area 1	3	0	2.92	2.99	2.96	2.96	0.04	1
		Reference Area 2	3	0	3.45	3.88	3.64	3.66	0.22	6
		Reference Area 3	3	0	3.02	3.56	3.20	3.26	0.27	9
		Reference Area 4	1	0	3.23	3.23	3.23	3.23	.	
		Reference Means					3.26	3.28		
		Study Area North	5	0	2.45	3.01	2.53	2.70	0.28	11
		Study Area South	5	0	2.40	4.12	3.01	3.05	0.70	23
		Study Means					2.77	2.88		
Mercury	2004	Reference Area 1	3	0	0.060	0.100	0.080	0.080	0.020	25
		Reference Area 2	3	0	0.060	0.100	0.070	0.080	0.020	27
		Reference Area 3	2	0	0.090	0.100	0.100	0.100	0.010	7
		Reference Area 4	3	0	0.090	0.110	0.100	0.100	0.010	10
		Reference Means					0.090	0.090		
		Study Area	10	0	0.050	0.150	0.090	0.090	0.030	30
	2005	Reference Area 1	3	0	0.130	0.180	0.180	0.160	0.030	16
		Reference Area 2	3	0	0.050	0.170	0.130	0.120	0.060	47
		Reference Area 3	3	0	0.080	0.160	0.140	0.130	0.040	30
		Reference Area 4	1	0	0.200	0.200	0.200	0.200	.	
		Reference Means					0.160	0.150		
		Study Area North	5	0	0.100	0.120	0.100	0.100	0.010	9
		Study Area South	5	0	0.050	0.110	0.070	0.080	0.020	35
		Study Means					0.090	0.090		
Selenium	2004	Reference Area 1	3	0	0.70	0.80	0.70	0.73	0.06	8
		Reference Area 2	3	0	0.50	0.80	0.80	0.70	0.17	25
		Reference Area 3	2	0	0.70	0.70	0.70	0.70	0.00	0
		Reference Area 4	3	0	0.70	0.80	0.80	0.77	0.06	8
		Reference Means					0.75	0.73		
		Study Area	10	0	0.50	0.80	0.60	0.66	0.11	16
	2005	Reference Area 1	3	0	0.56	0.67	0.60	0.61	0.06	9
		Reference Area 2	3	0	0.53	0.65	0.55	0.58	0.06	12
		Reference Area 3	3	1	<0.5	0.62	0.61			
		Reference Area 4	1	0	0.54	0.54	0.54	0.54	.	
		Reference Means					0.58			
		Study Area North	5	0	0.58	0.69	0.65	0.64	0.05	7
		Study Area South	5	1	<0.5	0.59	0.53			
		Study Means					0.59			

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
Silver	2004	Reference Area 1	3	0	0.140	0.260	0.180	0.190	0.060	32
		Reference Area 2	3	0	0.150	0.250	0.200	0.200	0.050	25
		Reference Area 3	2	0	0.160	0.160	0.160	0.160	0.000	0
		Reference Area 4	3	0	0.210	0.270	0.230	0.240	0.030	13
		Reference Means					0.190	0.200		
	Study Area	10	0	0.150	0.250	0.200	0.210	0.030	15	
	2005	Reference Area 1	3	1	<0.12	0.150	0.140			
		Reference Area 2	3	1	<0.12	0.140	0.130			
		Reference Area 3	3	1	<0.12	0.250	0.240			
		Reference Area 4	1	0	0.220	0.220	0.220	0.220		
		Reference Means					0.180			
		Study Area North	5	0	0.130	0.190	0.150	0.160	0.020	15
		Study Area South	5	0	0.130	0.250	0.210	0.200	0.040	21
		Study Means					0.180	0.180		
Strontium	2004	Reference Area 1	3	0	5.20	10.00	9.30	8.17	2.59	32
		Reference Area 2	3	0	8.90	13.00	10.00	10.60	2.12	20
		Reference Area 3	2	0	6.20	15.00	10.60	10.60	6.22	59
		Reference Area 4	3	0	5.10	18.00	6.00	9.70	7.20	74
		Reference Means					8.98	9.78		
	Study Area	10	0	4.40	18.00	8.95	10.00	4.64	46	
	2005	Reference Area 1	3	0	6.10	8.60	6.90	7.20	1.28	19
		Reference Area 2	3	0	6.80	26.80	10.90	14.83	10.56	97
		Reference Area 3	3	0	14.70	20.10	16.00	16.93	2.82	18
		Reference Area 4	1	0	8.40	8.40	8.40	8.40		
		Reference Means					10.55	11.84		
		Study Area North	5	0	5.60	14.20	6.10	8.02	3.60	59
		Study Area South	5	0	9.70	21.00	12.80	14.16	5.01	39
		Study Means					9.45	11.09		
Zinc	2004	Reference Area 1	3	0	31.00	32.00	31.00	31.30	0.58	2
		Reference Area 2	3	0	23.00	30.00	23.00	25.30	4.04	16
		Reference Area 3	2	0	27.00	30.00	28.50	28.50	2.12	7
		Reference Area 4	3	0	31.00	35.00	33.00	33.00	2.00	6
		Reference Means					28.88	29.50		
	Study Area	10	0	17.00	33.00	30.50	28.20	4.78	17	
	2005	Reference Area 1	3	0	25.60	32.40	29.30	29.10	3.40	12
		Reference Area 2	3	0	17.50	28.90	23.00	23.13	5.70	25
		Reference Area 3	3	0	24.70	30.90	27.10	27.57	3.13	12
		Reference Area 4	1	0	27.30	27.30	27.30	27.30		
		Reference Means					26.68	26.78		
		Study Area North	5	0	25.00	30.60	27.30	27.74	2.09	8
		Study Area South	5	0	21.20	26.20	24.60	24.10	1.89	8
		Study Means					25.95	25.92		
% Fat	2004	Reference Area 1	2	1	<0.5	0.70	0.60			
		Reference Area 2	3	0	0.50	1.90	1.10	1.17	0.70	60
		Reference Area 3	2	0	0.60	1.30	0.95	0.95	0.49	52
		Reference Area 4	3	0	0.60	0.70	0.60	0.63	0.06	9
		Reference Means					0.81			
	Study Area	10	2	<0.5	1.40	0.70				
	2005	Reference Area 1	3	0	0.50	0.60	0.60	0.57	0.06	10
		Reference Area 2	2	0	0.50	1.00	0.75	0.75	0.35	47
		Reference Area 3	2	0	0.50	0.70	0.60	0.60	0.14	23
		Reference Area 4	1	0	0.50	0.50	0.50	0.50		
		Reference Means					0.61	0.60		
		Study Area North	5	2	<0.5	0.60	0.50			
		Study Area South	5	0	0.50	2.20	0.60	0.92	0.72	120
		Study Means					0.55			
% Moisture	2004	Reference Area 1	3	0	78.00	81.00	79.00	79.30	1.53	2
		Reference Area 2	3	0	80.00	85.00	81.00	82.00	2.65	3
		Reference Area 3	2	0	79.00	82.00	80.50	80.50	2.12	3
		Reference Area 4	3	0	78.00	80.00	78.00	78.70	1.15	1
		Reference Means					79.63	80.10		
		Study Area	10	0	80.00	85.00	81.00	81.70	1.77	2



Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
% Moisture	2005	Reference Area 1	3	0	78.00	82.00	80.00	80.00	2.00	3
		Reference Area 2	3	0	78.00	83.00	81.00	80.67	2.52	3
		Reference Area 3	3	0	79.00	80.00	80.00	79.67	0.58	1
		Reference Area 4	1	0	80.00	80.00	80.00	80.00	.	
		Reference Means					80.25	80.08		
		Study Area North	5	0	80.00	81.00	80.00	80.20	0.45	1
		Study Area South	5	0	79.00	82.00	80.00	80.40	1.14	1
Study Means						80.00	80.30			

Note: - All units are mg/kg except where indicated  
 - Statistics are reported to one more significant digit than what is given for EQL (see Table 6-4)

### Analyses of 2005 Data

The first step in analyses of 2005 crab body burden data was to conduct PCA on log-transformed concentrations of eight frequently detected metals (arsenic, boron, copper, mercury, selenium, silver, strontium, zinc). The PCA included 2004 as well as 2005 data, since Metals PC1 scores were compared between the two years (see below). PC1 was positively correlated with concentrations of all metals except boron and strontium, and accounted for 38% of total variance (Table 6-15). Boron was uncorrelated with, and strontium negatively correlated with, PC1. PC2 was positively correlated with concentrations of mercury and boron, and negatively correlated with concentrations of copper, silver and strontium. PC3 was positively correlated with concentrations of boron and silver.

**Table 6-15 Correlations (Parametric or Pearson  $r$ ) Between Metal Concentrations in Crab Claw Composites and Principal Components Derived from Those Concentrations (2004, 2005)**

Metal	Correlation ( $r$ ) with:		
	PC1	PC2	PC3
Zinc	<b>0.871</b>	0.290	-0.124
Arsenic	<b>0.795</b>	-0.033	0.136
Selenium	<b>0.769</b>	-0.253	-0.199
Copper	<b>0.549</b>	<b>-0.657</b>	0.205
Silver	0.453	-0.445	<b>0.523</b>
Mercury	0.414	<b>0.787</b>	0.193
Boron	-0.120	0.416	<b>0.797</b>
Strontium	<b>-0.607</b>	-0.421	0.298
% variance	38	22	14

Notes: - Metals are listed in descending order of their correlations with PC1  
 -  $|r| \geq 0.5$  in bold  
 - Metal concentrations were  $\log_{10}$  transformed prior to deriving PC  
 -  $n = 41$  composites (21 from 2004; 20 from 2005)

Metals PC1 scores were used as a summary measure of total metals concentrations (excluding strontium and boron) for subsequent analyses. The positive correlations with PC1 for most metals indicated that higher concentrations of these metals tended to co-occur. The negative correlation between PC1 (and most metals) and strontium may indicate that strontium competes with other metals for binding sites in the claw and possibly other tissues or, more generally, that strontium “behaves differently”.

Metals PC2 and PC3 scores were not retained for further analyses for several reasons. First, PC2 and PC3 combined accounted for less variance than PC1. Second, secondary PCs may reflect some real differences in metal mixtures but could also result from non-

linearities in relationships identified by PC1, or the limited number of values and significant digits (never more than for EQL) for metals occurring at low concentrations. For example, the predicted value of Metal A based on concentrations of other metals might be 5.5, but observed values would be reported to only one significant digit if EQL = 1. Thus, observed values of 5 or 6 would agree with the predicted value to one significant digit. However, the difference between the observed and predicted values would be approximately 10%, which might be significant relative to other secondary sources of variance. For the crab PCA, correlations between the secondary axes and boron and silver should be regarded with caution, since these metals varied over a narrow range and PCA results would also depend on how values below EQL were treated. Third, PC2 scores were positively correlated with PC1 scores for 2005 samples (Figure 6-4), so results for analyses of the two PC would be similar (i.e., PC2 is redundant). The absence of a similar correlation in 2004 may suggest some mixture differences, real or analytical, between the two sample years. PC2 and PC3 were useful for identifying individual metals of potential interest. For example, mercury was strongly correlated with PC2, and if present as methyl mercury (i.e., the organic form) would be of interest for human health reasons and would be expected to behave differently than other metals.

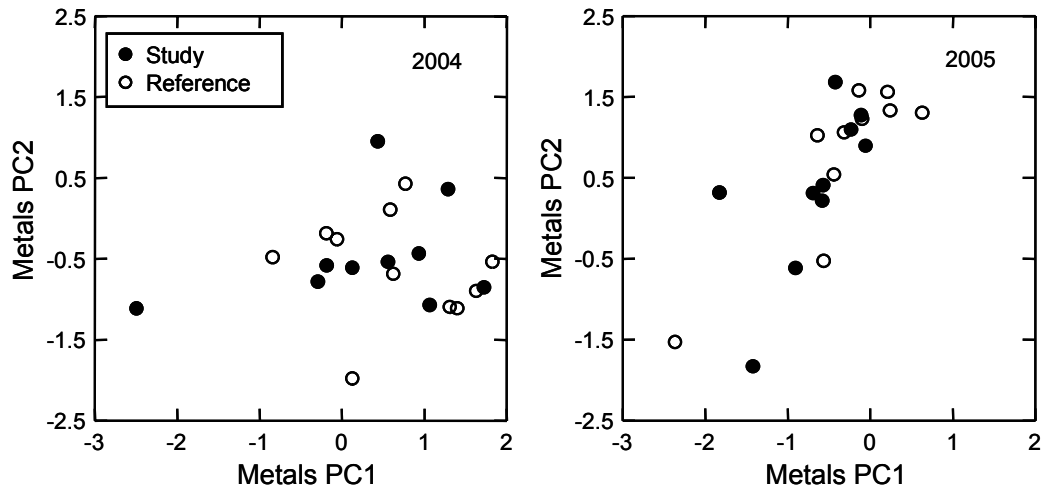


Figure 6-4 Metals PC2 versus PC1 Scores for Crab Claw Composites (2004, 2005)

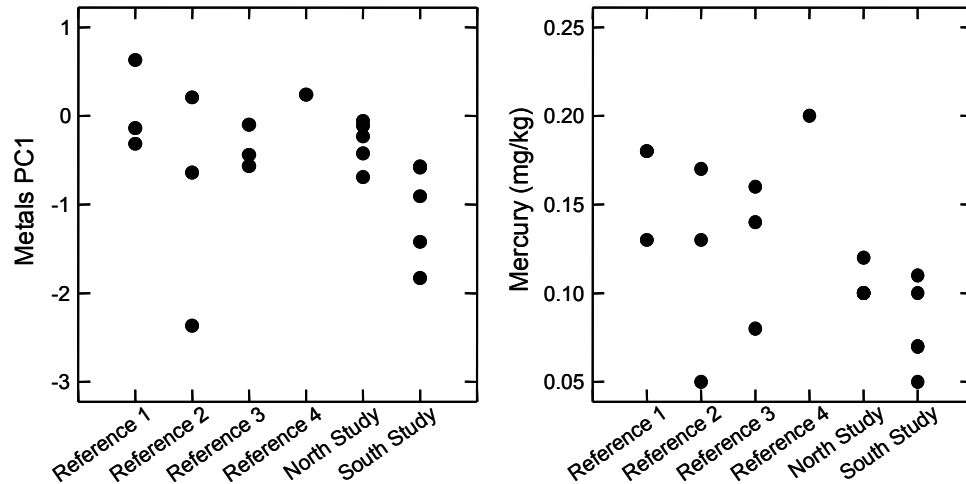
Moisture and fat content did not differ significantly among Reference Areas, between the Northern and Southern portion of the Study Area, or between the Study versus Reference Areas (Table 6-16), although fat content was lower in composites from the Northern portion of the Study Area than in composites from the Southern portion (Table 6-14).

**Table 6-16 Results of Modified Nested ANOVA Comparing Crab Body Burden Variables Among Areas (2005)**

Variable	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
% moisture	0.679	0.754	<i>0.824</i>	0.687	<i>0.773</i>
% fat (rank)	0.947	0.014	<i>0.076</i>	0.270	<i>0.731</i>
Metals PC1 (rank)	0.265	0.180	<b>0.028</b>	0.355	<i>0.171</i>
Arsenic	0.568	0.247	<i>0.235</i>	0.113	<i>0.056</i>
Boron	0.070	<i>0.452</i>	0.118	<i>0.882</i>	0.766
Copper	0.166	<i>0.461</i>	0.216	<i>0.276</i>	0.052
Mercury	0.226	<i>0.471</i>	0.271	<i>0.155</i>	0.013
Selenium	0.672	0.043	<b>0.010</b>	1.000	<i>0.960</i>
Silver	0.068	<i>0.427</i>	0.095	<i>0.542</i>	0.212
Strontium	0.095	<i>0.388</i>	0.089	<i>0.687</i>	0.449
Zinc	0.089	<i>0.396</i>	0.090	<i>0.809</i>	0.646

Notes: - Reference Area 4 (1 composite) excluded  
 - MSE = variance among composites within Areas; MS(AR) = variance among Reference Areas  
 - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

Overall, differences in metal concentrations among Areas were small (i.e., less than two-fold except for boron and strontium) and rarely significant. Here and elsewhere, results for individual metals should be interpreted with some caution, since there were zero or near-zero variances within some Areas and also some outliers relative to these low variances. Results would be most robust for Metals PC1 (a summary measure) and individual metals occurring at concentrations well above (i.e., more than 5 to 10 times) EQL. Metals PC1 scores, and concentrations of most metals (e.g., mercury), were lower in composites from the Southern portion of the Study Area than in composites from the Northern portion of the Study Area and from the Reference Areas (Figure 6-5). Differences in Metals PC1 scores, and concentrations of individual metals, among Reference Areas were not significant at  $p \leq 0.05$  (Table 6-16). *p* values for the Among References contrast were  $< 0.10$  for several metals, but there was no consistency to the rank order of the Reference Areas for these metals (which might account for some of the variance reflected in Metals PC2 or PC3). For example, boron concentrations were lowest, but zinc concentrations highest, in Reference Area 1. Metal concentrations in the single Reference Area 4 sample were usually as high or higher than concentrations in other Reference Areas (Table 6-14; Figure 6-5). The Study versus Reference contrast was not significant for Metals PC1 or any individual metal when tested against the appropriate error as recommended by Quinn and Keough (2002). When *p* values for the contrast were low, concentrations were *lower* in the Study Area than in the Reference Areas (e.g., arsenic, copper, mercury). Finally, strontium concentrations in Reference Areas 2 and 3 were double those in other Areas (Table 6-14), a unique spatial pattern not observed for other metals.



**Figure 6-5 Distributions of Metals PC1 Scores and Mercury Concentrations for Crab Claw Composites (2005)**

Note: - Some points may represent more than one composite

Moisture content was not significantly correlated with any biological or body burden variables (Table 6-17). The strongest correlation (negative) was with fat content, although fat accounted for less than 10% of the dry weight content (100-% moisture) of the claw tissue. Fat content was significantly negatively correlated, and Metals PC1 significantly positively correlated, with size (carapace width, claw height). Mercury concentrations were significantly positively correlated, and strontium concentrations significantly negatively correlated, with size, as expected based on correlations between these two metals and PC1. Thus, larger crab had lower fat content and higher concentrations of most metals than smaller crab. The larger crab may have been converting fat to other body tissue rather than using fat for energy storage, although the claw would not be a major site of fat storage. The positive correlation between metal concentrations and size may indicate that some biomagnification or increase in metal concentrations with size and age occurred (possible for mercury and selenium, but not expected for most other metals; Newman and Unger 2003). Alternatively, the correlations with size may be a function of physiological differences affecting uptake (e.g., changes in gill surface area: body weight with size). Frequencies of recent moult were uncorrelated with body burden variables. Correlations among Metals PC1, mercury and strontium were as expected, based on the PCA results in Table 6-15.

**Table 6-17 Spearman Rank Correlations (rs) Among Crab Body Burden Variables, and Between Those Variables and Biological Characteristics (2005)**

	% moisture	% fat	Metals PC1	Mercury	Strontium
Carapace width	0.126	<b>-0.538*</b>	<b>0.693**</b>	<b>0.477*</b>	<b>-0.737**</b>
Claw height	0.088	<b>-0.559*</b>	<b>0.709**</b>	<b>0.477*</b>	<b>-0.726**</b>
% recent moult	-0.278	-0.225	0.280	-0.018	-0.071
% moisture		-0.426	-0.159	-0.182	-0.090
% fat			-0.370	0.053	0.328
Metals PC1				<b>0.728**</b>	<b>-0.675**</b>
Mercury					<b>-0.479*</b>

Notes: - Reference Area 4 (1 composite of 8 crab) excluded  
 - n = 19 composites, except for % fat (n = 17 composites)  
 - \*p ≤ 0.05; \*\*p ≤ 0.01; rs at p ≤ 0.05 in bold

### Comparison of 2005 versus 2004 Results

Results of RM ANOVA (Table 6-7) comparing crab body burden variables between Areas and years (2004 and 2005) are provided in Table 6-18. The tests had limited power, especially with Reference Area 4 excluded, so  $p \leq 0.10$  (in bold in Table 6-18) instead of the traditional  $p \leq 0.05$  was used to define statistical significance. Even at  $p \leq 0.10$ , few terms or tests were significant. Many differences over time or space were small because Area means rather than values from individual composites were analyzed. With Reference Area 4 excluded, the RM ANOVA was not estimable for selenium because the differences between 2004 and 2005 were identical for the other three References (i.e., Year x Among References or Within Areas error variance was 0).

**Table 6-18 Results of Repeated Measures ANOVA Comparing Crab Body Burden Variables Among Areas and Between Years (2004, 2005)**

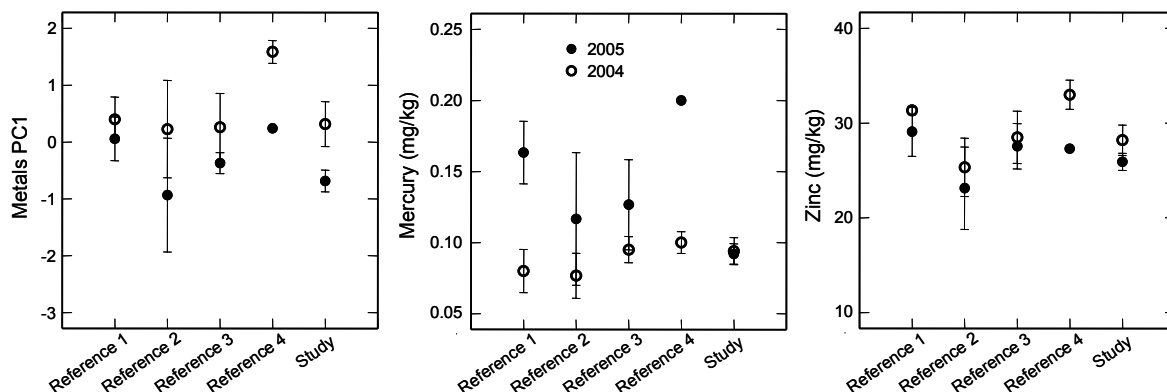
Variable	Between Areas		Within Areas		Between Areas		Within Areas	
	Study versus References (SR)	Among References	Year	Year x SR	Study versus References (SR)	Among References	Year	Year x SR
% moisture	0.427	0.297	0.378	0.403	0.589	0.265	0.255	0.532
% fat	0.950	0.135	0.282	0.387	0.879	0.230	0.336	0.422
Metals PC1	0.586	<b>0.099</b>	<b>0.037</b>	0.822	0.745	0.338	<b>0.071</b>	0.611
Arsenic	0.378	0.164	0.117	0.580	0.567	0.742	0.156	0.367
Boron	0.869	0.708	0.204	0.772	0.922	0.629	0.340	0.916
Copper	0.658	0.200	<b>0.060</b>	0.693	0.811	0.142	<b>0.085</b>	0.440
Mercury	0.367	0.300	0.194	0.174	0.367	0.571	0.261	0.236
Selenium	0.239	0.779	<b>0.032</b>	0.269	NE	NE	NE	NE
Silver	0.837	0.398	0.773	1.000	0.187	0.905	0.537	0.973
Strontium	0.944	0.252	0.515	0.828	0.845	0.249	0.430	0.672
Zinc	0.749	<b>0.066</b>	0.115	0.845	0.913	<b>0.015</b>	<b>0.042</b>	0.624

Notes: - See Table 6-7 and Section 6.3.2.1 for further explanation of the RM ANOVA  
 - NE = Not Estimable (Within Areas error variance = 0)

$p$  values for the Between Areas Study versus Reference (SR), and the Within Areas Year x SR, terms were all  $> 0.1$  and often  $> 0.5$  (Table 6-18), indicating that:

- averages over both years did not differ significantly between the Study and Reference Areas (Between Areas SR)
- any small differences that occurred did not change between years (Within Areas SR)

Moisture and fat content did not vary significantly over either time or space. Metals PC1 scores were greater in 2004 than in 2005 in every Area (Figure 6-6), with  $p \leq 0.10$  for the Year term. Decreases in metal concentrations from 2004 to 2005 were most significant for copper, selenium, and zinc (lowest  $p$  for year in Table 6-18; zinc concentrations are plotted in Figure 6-6). Concentrations of other metals (e.g. mercury in Figure 6-6) either did not change over time or were greater in 2005 in most Areas. The differences in temporal changes among metals (i.e., with some increasing and others remaining the same or decreasing) account for some of the secondary variance in metal mixtures identified by Metals PC2 and PC3 (Table 6-15).



**Figure 6-6 Metals PC1 Scores, Mercury and Zinc Concentrations for Crab Claw Composites (2004, 2005)**

Note: - Values are Area means ± 1 SE (vertical bars); SE were based on variance among composites

**6.4.2.2 Plaice**

**Liver**

Summary statistics for detected substances in plaice liver in 2004 and 2005 are provided in Table 6-19. Raw data for 2005 are provided in Appendix C-2. HCs detected in the >C<sub>10</sub>-C<sub>21</sub> and >C<sub>21</sub>-C<sub>32</sub> range in both 2004 and 2005 showed no resemblance to drill fluid. HC peaks observed on chromatograms for liver (Appendix C-2; also see Husky Energy 2005 for chromatograms for 2004 samples) were consistent with those expected for extracted fatty acid compounds (Maxxam Analytics, pers. comm.).

**Table 6-19 Summary Statistics for Plaice Liver Body Burden (2004, 2005)**

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV%	
>C <sub>10</sub> -C <sub>21</sub>	2004	Reference Area 1	3	0	31.0	87.0	70.0	62.7	28.7	46	
		Reference Area 2	3	0	44.0	56.0	49.0	49.7	6.0	12	
		Reference Area 3	3	0	76.0	85.0	78.0	79.7	4.7	6	
		Reference Area 4	3	0	110.0	150.0	140.0	133.3	20.8	16	
		Reference Means					84.3	81.3			
		Study Area	9	0	47.0	110.0	65.0	74.3	24.7	33	
	2005	Reference Area 1	3	0	33.0	120.0	81.0	78.0	43.6	56	
		Reference Area 2	3	0	44.0	73.0	44.0	53.7	16.7	31	
		Reference Area 3	3	0	56.0	68.0	63.0	62.3	6.0	10	
		Reference Area 4	3	0	99.0	130.0	100.0	109.7	17.6	16	
		Reference Means					72.0	75.9			
		Study Area North	5	0	42.0	110.0	61.0	67.0	25.7	38	
		Study Area South	5	0	35.0	74.0	53.0	55.0	14.2	26	
		Study Means					57.0	61.0			
>C <sub>21</sub> -C <sub>32</sub>	2004	Reference Area 1	3	0	62.0	130.0	79.0	90.3	35.4	39	
		Reference Area 2	3	0	64.0	110.0	71.0	81.7	24.8	30	
		Reference Area 3	3	0	57.0	100.0	65.0	74.0	22.9	31	
		Reference Area 4	3	0	56.0	96.0	91.0	81.0	21.8	27	
		Reference Means					76.5	81.8			
		Study Area	9	0	40.0	120.0	55.0	62.1	23.3	37	
	2005	Reference Area 1	3	0	43.0	57.0	46.0	48.7	7.4	15	
		Reference Area 2	3	0	45.0	73.0	63.0	60.3	14.2	24	
		Reference Area 3	3	0	47.0	75.0	56.0	59.3	14.3	24	
		Reference Area 4	3	0	60.0	110.0	67.0	79.0	27.1	34	
		Reference Means					58.0	61.8			
		Study Area North	5	0	42.0	81.0	70.0	65.6	16.1	25	
		Study Area South	5	0	50.0	93.0	71.0	69.2	16.5	24	
		Study Means									

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV%
>C <sub>21</sub> -C <sub>32</sub>	2005	Study Means					70.5	67.4		
Arsenic	2004	Reference Area 1	3	0	2.80	4.30	2.90	3.33	0.84	25
		Reference Area 2	3	0	1.80	5.20	4.00	3.67	1.72	47
		Reference Area 3	3	0	3.10	3.40	3.10	3.20	0.17	5
		Reference Area 4	3	0	4.10	5.40	4.30	4.60	0.70	15
		Reference Means					3.58	3.70		
	Study Area	10	0	1.80	5.80	3.35	3.42	1.08	32	
	2005	Reference Area 1	3	0	3.11	4.49	3.40	3.67	0.73	20
		Reference Area 2	3	0	2.98	3.26	3.25	3.16	0.16	5
		Reference Area 3	3	0	3.90	4.92	4.39	4.40	0.51	12
		Reference Area 4	3	0	1.74	3.21	2.58	2.51	0.74	29
		Reference Means					3.41	3.44		
		Study Area North	5	0	2.60	3.79	2.83	3.12	0.54	17
		Study Area South	5	0	2.54	8.27	3.43	4.17	2.36	57
		Study Means					3.13	3.65		
Cadmium	2004	Reference Area 1	3	0	0.380	0.690	0.410	0.490	0.170	35
		Reference Area 2	3	0	0.460	0.650	0.480	0.530	0.100	20
		Reference Area 3	3	0	0.380	0.410	0.410	0.400	0.020	4
		Reference Area 4	3	0	0.490	0.650	0.530	0.560	0.080	15
		Reference Means					0.460	0.500		
	Study Area	10	0	0.330	0.540	0.435	0.440	0.070	17	
	2005	Reference Area 1	3	0	0.362	0.766	0.538	0.560	0.200	36
		Reference Area 2	3	0	0.401	0.575	0.550	0.510	0.090	18
		Reference Area 3	3	0	0.573	0.805	0.601	0.660	0.130	19
		Reference Area 4	3	0	0.283	0.518	0.336	0.380	0.120	33
		Reference Means					0.506	0.530		
		Study Area North	5	0	0.291	0.532	0.461	0.420	0.100	24
		Study Area South	5	0	0.377	0.723	0.414	0.460	0.150	32
		Study Means					0.438	0.440		
Copper	2004	Reference Area 1	3	0	3.10	4.90	4.20	4.07	0.91	22
		Reference Area 2	3	0	2.80	4.60	4.50	3.97	1.01	26
		Reference Area 3	3	0	3.30	4.70	4.00	4.00	0.70	18
		Reference Area 4	3	0	3.00	6.60	5.10	4.90	1.81	37
		Reference Means					4.45	4.23		
	Study Area	10	0	1.80	6.00	3.40	3.62	1.42	39	
	2005	Reference Area 1	3	0	2.88	5.74	5.25	4.62	1.53	33
		Reference Area 2	3	0	2.60	6.92	3.75	4.42	2.24	51
		Reference Area 3	3	0	4.57	5.46	4.88	4.97	0.45	9
		Reference Area 4	3	0	3.83	4.28	3.95	4.02	0.23	6
		Reference Means					4.46	4.51		
		Study Area North	5	0	1.69	4.58	2.29	2.95	1.33	45
		Study Area South	5	0	3.15	7.22	4.74	4.80	1.59	33
		Study Means					3.52	3.87		
Iron	2004	Reference Area 1	3	0	22.0	66.0	44.0	44.0	22.0	50
		Reference Area 2	3	0	36.0	58.0	52.0	48.7	11.4	23
		Reference Area 3	3	0	30.0	36.0	33.0	33.0	3.0	9
		Reference Area 4	3	0	32.0	45.0	42.0	39.7	6.8	17
		Reference Means					42.8	41.3		
	Study Area	10	0	29.0	52.0	41.5	40.5	7.8	19	
	2005	Reference Area 1	3	0	40.0	57.0	45.0	47.3	8.7	18
		Reference Area 2	3	0	37.0	52.0	41.0	43.3	7.8	18
		Reference Area 3	3	0	52.0	70.0	64.0	62.0	9.2	15
		Reference Area 4	3	0	32.0	67.0	33.0	44.0	19.9	45
		Reference Means					45.8	49.2		
		Study Area North	5	0	29.0	111.0	36.0	54.6	34.2	63
		Study Area South	5	0	32.0	55.0	42.0	44.2	9.0	20
		Study Means					39.0	49.4		
Manganese	2004	Reference Area 1	3	0	0.70	0.80	0.80	0.77	0.06	8
		Reference Area 2	3	0	0.80	0.90	0.80	0.83	0.06	7
		Reference Area 3	3	0	0.80	1.00	0.80	0.87	0.12	13
		Reference Area 4	3	0	0.70	1.00	0.90	0.87	0.15	18
		Reference Means					0.83	0.83		
	Study Area	10	0	0.70	1.00	0.80	0.83	0.09	11	
	2005	Reference Area 1	3	0	0.85	0.92	0.92	0.90	0.04	5

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV%
Manganese	2005	Reference Area 2	3	0	0.99	1.59	1.07	1.22	0.33	27
		Reference Area 3	3	0	0.76	0.86	0.81	0.81	0.05	6
		Reference Area 4	3	0	0.72	0.94	0.81	0.82	0.11	13
		Reference Means					0.90	0.94		
		Study Area North	5	0	0.71	0.96	0.86	0.86	0.11	12
		Study Area South	5	0	0.67	0.91	0.89	0.84	0.10	12
		Study Means					0.88	0.85		
Mercury	2004	Reference Area 1	3	0	0.020	0.040	0.030	0.030	0.010	33
		Reference Area 2	3	0	0.030	0.040	0.040	0.040	0.010	16
		Reference Area 3	3	0	0.020	0.040	0.030	0.030	0.010	33
		Reference Area 4	3	0	0.030	0.040	0.030	0.030	0.010	17
		Reference Means					0.033	0.030		
		Study Area	10	0	0.020	0.040	0.030	0.030	0.000	16
		Study Means								
	2005	Reference Area 1	3	0	0.040	0.050	0.040	0.040	0.010	13
		Reference Area 2	3	0	0.020	0.060	0.050	0.040	0.020	48
		Reference Area 3	3	0	0.050	0.060	0.050	0.050	0.010	11
		Reference Area 4	3	0	0.030	0.030	0.030	0.030	0.000	0
		Reference Means					0.043	0.040		
		Study Area North	5	0	0.020	0.040	0.020	0.030	0.010	39
		Study Area South	5	0	0.020	0.040	0.030	0.030	0.010	24
		Study Means					0.025	0.030		
Selenium	2004	Reference Area 1	3	0	1.70	2.10	1.90	1.90	0.20	11
		Reference Area 2	3	0	2.10	2.50	2.40	2.33	0.21	9
		Reference Area 3	3	0	1.90	2.20	2.00	2.03	0.15	8
		Reference Area 4	3	0	1.30	1.80	1.70	1.60	0.26	17
		Reference Means					2.00	1.97		
		Study Area	10	0	1.70	2.30	1.95	1.98	0.18	9
		Study Means								
	2005	Reference Area 1	3	0	1.88	2.36	2.09	2.11	0.24	11
		Reference Area 2	3	0	2.05	2.34	2.30	2.23	0.16	7
		Reference Area 3	3	0	2.51	2.61	2.53	2.55	0.05	2
		Reference Area 4	3	0	1.44	1.86	1.73	1.68	0.22	13
		Reference Means					2.16	2.14		
		Study Area	5	0	1.74	2.43	2.28	2.17	0.28	13
Silver	2004	Reference Area 1	3	3	<0.12	<0.12	<0.12			
		Reference Area 2	3	3	<0.12	<0.12	<0.12			
		Reference Area 3	3	2	<0.12	0.130	<0.12			
		Reference Area 4	3	2	<0.12	0.180	<0.12			
		Reference Means								
		Study Area	10	10	<0.12	<0.12	<0.12			
	2005	Reference Area 1	3	3	<0.12	<0.12	<0.12			
		Reference Area 2	3	3	<0.12	<0.12	<0.12			
		Reference Area 3	3	1	<0.12	0.300	0.140			
		Reference Area 4	3	3	<0.12	<0.12	<0.12			
Strontium	2004	Reference Area 1	3	3	<1.5	<1.5	<1.5			
		Reference Area 2	3	3	<1.5	<1.5	<1.5			
		Reference Area 3	3	3	<1.5	<1.5	<1.5			
		Reference Area 4	3	2	<1.5	1.60	<1.5			
		Reference Means								
		Study Area	10	10	<1.5	<1.5	<1.5			
		Study Area North	5	4	<0.12	0.250	<0.12			
		Study Area South	5	4	<0.12	0.120	<0.12			
Uranium	2005	Reference Area 1	3	3	<0.02	<0.02	<0.02			
		Reference Area 2	3	2	<0.02	0.022	<0.02			
		Reference Area 3	3	3	<0.02	<0.02	<0.02			
		Reference Area 4	3	3	<0.02	<0.02	<0.02			
		Reference Means								
		Study Area North	5	5	<0.02	<0.02	<0.02			
		Study Area South	5	5	<0.02	<0.02	<0.02			
		Study Means								



Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV%
Zinc	2004	Reference Area 1	3	0	23.00	25.00	23.00	23.67	1.15	5
		Reference Area 2	3	0	23.00	24.00	24.00	23.67	0.58	2
		Reference Area 3	3	0	22.00	26.00	22.00	23.33	2.31	10
		Reference Area 4	3	0	22.00	29.00	28.00	26.33	3.79	14
		Reference Means					24.25	24.25		
		Study Area	10	0	19.00	24.00	22.50	22.20	1.75	8
	2005	Reference Area 1	3	0	23.20	30.30	27.00	26.83	3.55	13
		Reference Area 2	3	0	25.50	27.80	27.50	26.93	1.25	5
		Reference Area 3	3	0	24.90	28.10	26.90	26.63	1.62	6
		Reference Area 4	3	0	22.60	27.10	22.90	24.20	2.52	10
		Reference Means					26.08	26.15		
		Study Area North	5	0	20.00	27.20	21.60	23.18	3.36	15
		Study Area South	5	0	21.70	28.70	25.10	25.26	2.70	11
		Study Means					23.35	24.22		
% Fat	2004	Reference Area 1	3	0	14.00	23.00	15.00	17.33	4.93	28
		Reference Area 2	3	0	11.00	13.00	12.00	12.00	1.00	8
		Reference Area 3	3	0	11.00	17.00	14.00	14.00	3.00	21
		Reference Area 4	3	0	15.00	18.00	16.00	16.33	1.53	9
		Reference Means					14.25	14.92		
		Study Area	10	0	10.00	20.00	12.50	13.30	2.87	22
	2005	Reference Area 1	3	0	11.00	14.00	13.00	12.67	1.53	12
		Reference Area 2	2	0	14.00	15.00	14.50	14.50	0.71	5
		Reference Area 3	3	0	12.00	17.00	13.00	14.00	2.65	19
		Reference Area 4	3	0	17.00	25.00	24.00	22.00	4.36	20
		Reference Means					16.13	15.79		
		Study Area North	2	0	<10	18.00	14.00	14.00	5.66	40
		Study Area South	5	0	13.00	21.00	18.00	17.20	3.19	19
		Study Means					16.00	15.60		
% Moisture	2004	Reference Area 1	3	0	63.00	70.00	68.00	67.00	3.61	5
		Reference Area 2	3	0	70.00	71.00	70.00	70.33	0.58	1
		Reference Area 3	3	0	66.00	71.00	68.00	68.33	2.52	4
		Reference Area 4	3	0	66.00	69.00	67.00	67.33	1.53	2
		Reference Means					68.25	68.25		
		Study Area	10	0	66.00	73.00	70.00	69.90	2.02	3
	2005	Reference Area 1	3	0	67.00	70.00	69.00	68.67	1.53	2
		Reference Area 2	3	0	61.00	68.00	66.00	65.00	3.61	6
		Reference Area 3	3	0	67.00	70.00	69.00	68.67	1.53	2
		Reference Area 4	3	0	59.00	65.00	61.00	61.67	3.06	5
		Reference Means					66.25	66.00		
		Study Area North	5	0	64.00	70.00	69.00	67.40	2.70	4
		Study Area South	5	0	61.00	69.00	64.00	64.40	2.97	5
		Study Means					66.50	65.90		

Note: - All units are mg/kg except where indicated  
 - Statistics are reported to one more significant digit than what is given for EQL (see Table 6-4)

### Analysis of 2005 Data

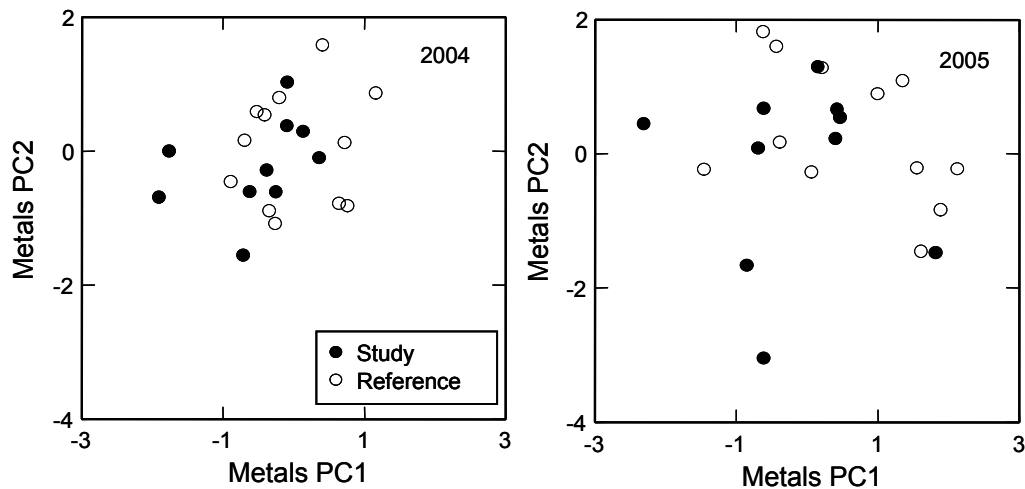
The first step in analyses of plaice liver body burdens was to conduct a PCA on log-transformed concentrations of eight metals (arsenic, cadmium, copper, iron, manganese, mercury, selenium, zinc). The PCA included all 2004 and 2005 samples since PC scores were compared between years (see below). Concentrations of all metals except iron and manganese were positively correlated with each other and with PC1 (Table 6-20), which served as a summary measure of total metal concentrations. PC2 and PC3 each accounted for approximately the same amount of variance, and combined, accounted for less variance than PC1. These minor axes were not used for subsequent analyses, for reasons given in Section 6.4.2.1, and because they were difficult to interpret. Specifically, correlations between iron and manganese versus PC2 were of opposite sign, but correlations with PC3 were of the same sign. Thus, the relationship between the two metals varied over space or time (or both). Overall, the PCA was useful in terms

of indicating that iron and manganese behaved differently than other metals. Based on the first two PCs, metal concentrations were more variable in 2005 than in 2004 (Figure 6-7).

**Table 6-20 Correlations (Parametric or Pearson r) Between Metal Concentrations in Plaiice Liver Composites and Principal Components (PC) Derived from Those Concentrations (2004, 2005)**

Metal	Correlation (r) with:		
	PC1	PC2	PC3
Cadmium	<b>0.759</b>	-0.375	-0.053
Zinc	<b>0.740</b>	0.445	0.112
Copper	<b>0.729</b>	0.354	-0.284
Mercury	<b>0.680</b>	0.113	0.040
Arsenic	<b>0.652</b>	-0.180	-0.438
Selenium	0.492	-0.230	<b>0.608</b>
Iron	0.254	<b>-0.665</b>	0.483
Manganese	0.015	<b>0.672</b>	<b>0.608</b>
% variance	36	18	16

- Notes:
- Metals are listed in descending order of their correlations with PC1
  - $|r| \geq 0.5$  in bold
  - Metal concentrations were  $\log_{10}$  transformed prior to deriving PC
  - $n = 44$  composites (22 from 2004; 22 from 2005)



**Figure 6-7 Metals PC2 versus PC1 Scores for Plaiice Liver Composites (2004, 2005)**

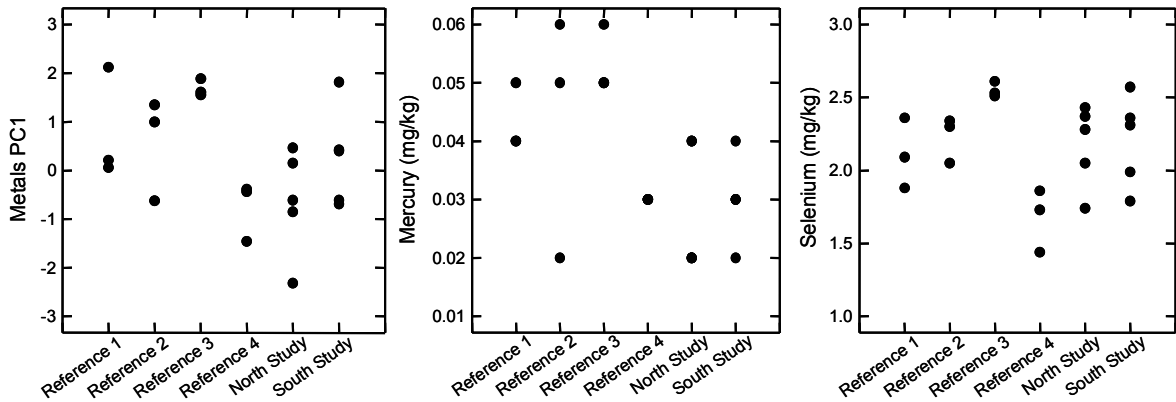
Moisture and fat content differed significantly among the four Reference Areas (Table 6-21). Moisture content was lower in Reference Area 4, and fat content was higher in Reference Area 3, than in other Reference Areas (Table 6-14). Moisture and fat content in the Northern and Southern portion of the Study Area were within the Reference range, and differences between the Northern and Southern portion of the Study Area and between Study versus Reference Areas were not significant.

**Table 6-21 Results of Modified Nested ANOVA Comparing Plaice Liver Body Burden Variables Among Areas (2005)**

Variable	p values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
% moisture	<b>0.017</b>	<i>0.476</i>	0.933	<i>0.971</i>	0.100
% fat	<b>0.020</b>	<i>0.634</i>	0.268	<i>0.963</i>	0.912
Metals PC1	<b>0.044</b>	<i>0.476</i>	0.154	<i>0.387</i>	0.082
Arsenic	0.351	0.315	<i>0.211</i>	<i>0.747</i>	<i>0.706</i>
Cadmium	0.120	<i>0.752</i>	0.609	<i>0.403</i>	0.163
Copper	0.872	0.024	<i>0.057</i>	0.119	<i>0.310</i>
Iron	0.633	0.357	<i>0.417</i>	0.974	<i>0.978</i>
Manganese	<b>0.010</b>	<i>0.944</i>	0.863	<i>0.575</i>	0.169
Mercury	0.087	<i>0.861</i>	0.762	<i>0.153</i>	0.007
Selenium	<b>0.005</b>	<i>0.944</i>	0.850	<i>0.871</i>	0.659
Zinc	0.578	0.242	<i>0.248</i>	0.140	<i>0.120</i>
>C <sub>10</sub> -C <sub>21</sub> HCs (rank)	0.162	<i>0.649</i>	0.491	<i>0.419</i>	0.210
>C <sub>21</sub> -C <sub>32</sub> HCs (rank)	0.300	0.800	<i>0.754</i>	0.412	<i>0.290</i>

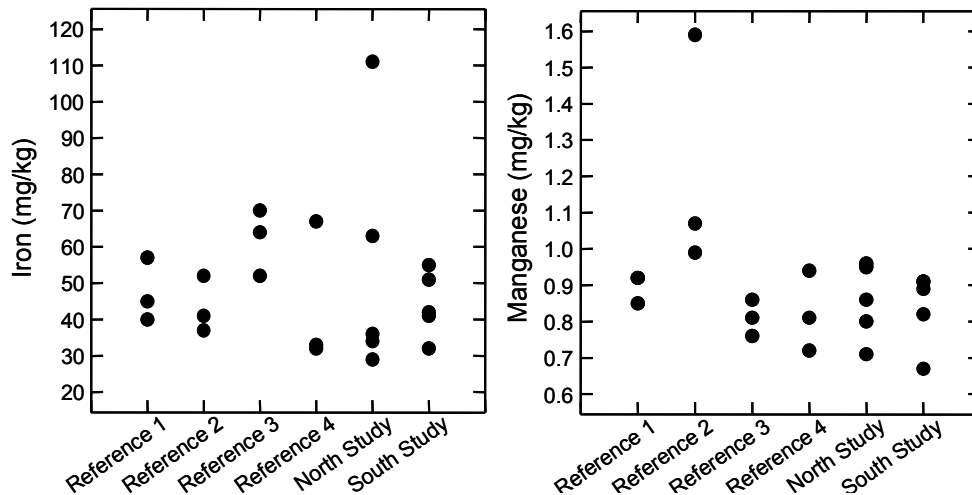
Notes: - MSE = variance among composites within Areas; MS(AR) = variance among Reference Areas  
 - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

Overall, metal concentrations differed more among the Reference Areas than between the Northern and Southern portion of the Study Area or between Study versus Reference Areas, although most differences among Areas were small and not significant for individual metals (Table 6-21). Metals PC1 scores differed significantly among the Reference Areas, with scores lowest in Reference Area 4 (Figure 6-8). Mercury and selenium followed that spatial pattern reasonably well (Figure 6-8); other metals, especially manganese and iron, did not. Manganese concentrations were higher in Reference Area 2, and iron concentrations higher in Reference Area 3, than in other Areas (Figure 6-9). Differences among Reference Areas were not significant for iron because of the high variances within some Areas. Differences in metal concentrations between the Northern and Southern portion of the Study Area and between the Study versus Reference Areas were never significant, and concentrations of all eight metals in the Study Area(s) were either within or below the Reference range.



**Figure 6-8 Distributions of Metals PC1 Scores, Mercury and Selenium Concentrations for Plaice Liver Composites (2005)**

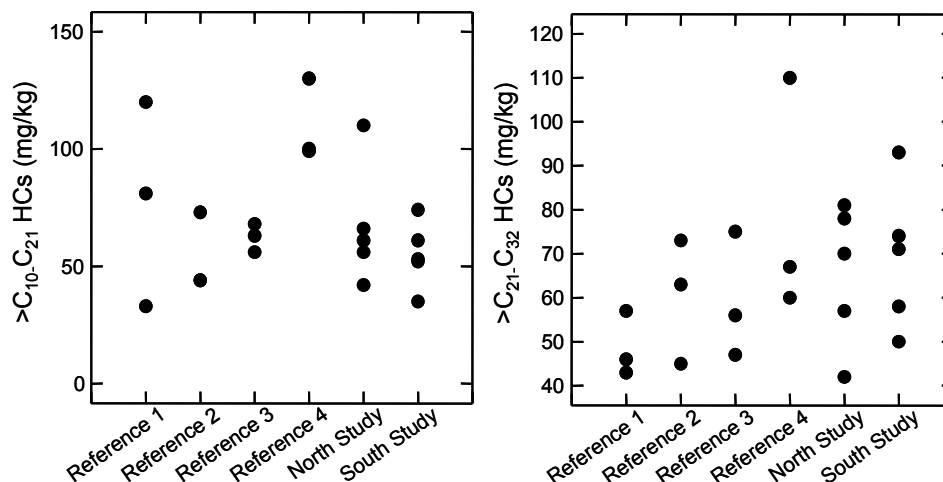
Note: - Some points may represent more than one composite



**Figure 6-9 Distributions of Iron and Manganese Concentrations for Plaice Liver Composites (2005)**

Note: - Some points may represent more than one composite

Concentrations of >C<sub>10</sub>-C<sub>21</sub> and >C<sub>21</sub>-C<sub>32</sub> HCs varied mostly within Areas, and none of the Area differences tested in the nested ANOVA were significant (Table 6-21, Figure 6-10).



**Figure 6-10 Distributions of >C<sub>10</sub>-C<sub>21</sub> and >C<sub>21</sub>-C<sub>32</sub> HC Concentrations for Plaiice Liver Composites (2005)**

Note: - Some points may represent more than one composite

Gutted weight (i.e., size), and especially the large difference in size and maturity status between Reference Area 3 and other Areas (Section 6.4.1.2), had relatively small effects on most liver body burden variables (Table 6-22). Metal PC1 scores (i.e., concentrations of most metals) were significantly positively correlated with gutted weight, potential evidence of biomagnification or changes in physiology and metal uptake/elimination rates associated with size differences. Metal concentrations in Reference Area 3 were similar to or greater than in other Areas (Figure 6-8). However, the correlation between size and Metals PC1 was still positive (0.411; 0.05 < *p* < 0.10) with Reference Area 3 excluded, indicating that the relatively small size differences within and among the other Areas accounted for most of the size “effects”.

**Table 6-22 Spearman Rank Correlations (*r<sub>s</sub>*) Among Plaiice Liver Burden Variables, and Between Those Variables and Composite Mean Gutted Weights (2005)**

	% moisture	% fat	Metals PC1	Iron	Manganese	>C <sub>10</sub> -C <sub>21</sub> HCs	>C <sub>21</sub> -C <sub>32</sub> HCs
Gutted weight	0.121	-0.109	<b>0.561**</b>	0.209	-0.405	-0.032	-0.069
% moisture		<b>-0.964**</b>	0.397	0.132	0.220	-0.207	<b>-0.473*</b>
% fat			<b>-0.427*</b>	-0.202	-0.341	0.395	<b>0.641**</b>
Metals PC1				0.265	-0.165	<b>-0.435*</b>	-0.141
Iron					<b>-0.486*</b>	-0.267	-0.410
Manganese						-0.006	-0.214
>C <sub>10</sub> -C <sub>21</sub> HCs							0.074

Notes: - *n* = 22 composites, except for % fat (*n* = 18 composites)  
 - \**p* ≤ 0.05; \*\* *p* ≤ 0.01; *r<sub>s</sub>* at *p* ≤ 0.05 in bold

Fat and moisture content were almost perfectly negatively correlated (Table 6-22), which confounded interpretation of correlations between these two variables and other body burden variables (see below). The strong negative correlation was expected and also observed in 2004 samples (Husky Energy 2005; *r<sub>s</sub>* between the two variables in 2004 was also -0.964). The liver is an important site for fat storage, and the fat content in liver

samples accounted for approximately 50% of the dry weight content (i.e., liver tissue is mostly water plus fat).

Metals PC1 scores were positively correlated with moisture content and negatively correlated with fat content (Table 6-22). When *dry* weight concentrations are positively correlated with moisture content, *wet* weight concentrations may be more similar among samples, or differ more among Areas than *dry* weight concentrations.

Concentrations of most metals should be uncorrelated with fat content, except that a positive correlation would be expected for lipophilic organometals such as methyl mercury. Therefore, the negative correlation between Metals PC1 and fat content was probably an artifact of the strong negative correlation between moisture and fat content. However, it is possible that fat rather than moisture content (or some correlate of both) has a more direct effect on metal uptake and body burdens. For example, plaice with a lower liver fat content may be “weaker”, “less healthy” and less able to regulate metal uptake and elimination.

Iron and manganese were included in Table 6-22 mostly to indicate that one or both metals may not behave like other metals (as summarized by Metals PC1). Any conclusions beyond that are probably unwarranted. Concentrations of the two metals were significantly negatively correlated in 2005, but not in 2004, when there was minimal variance of manganese concentrations.

Organic compounds such as HCs are more lipophilic than metals and concentrations should be greater when fat content is higher (i.e., HC concentrations should be positively correlated with fat content). Furthermore, if the HCs measured in plaice liver were primarily fatty acids (see Section 6.4.2.2), a positive correlation between HC concentrations and fat content would also be expected. Analytically, fatty acids can be included in measurements of both HCs and lipids (J. McDonald, pers. comm.). Many lipids are also derived from fatty acids, and one might expect a close correlation between concentrations of precursors (fatty acids) and their derivatives (lipids). In both 2005 (Table 6-22) and 2004 (Husky Energy 2005),  $>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HC concentrations in liver samples were positively correlated with fat content.

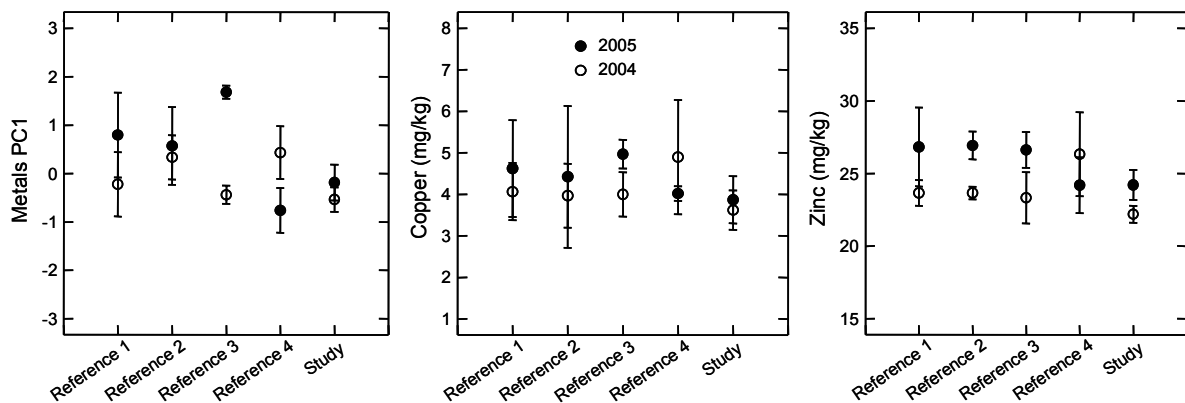
### ***Comparison of 2005 Versus 2004 Results***

Results of RM ANOVA comparing body burden results between 2005 and 2004 are provide in Table 6-23. Again, because the tests have limited power, a significance level of  $p \leq 0.10$  (in bold in Table 6-23) rather than  $p \leq 0.05$  was used. Moisture and fat content did not vary significantly over either space or time. There were no consistent differences among Areas in Metals PC1 scores over both years (Between Areas terms in Table 6-23; Figure 6-11). However, copper and zinc concentrations differed significantly between the Study versus Reference Areas (Table 6-23). Concentrations of both metals were somewhat lower in the Study Area than in Reference Areas in both years (Figure 6-11). The SR differences were small and significant only because consistent differences among the Reference Areas (i.e., the error variance for the SR contrast) were also small.

**Table 6-23 Results of Repeated Measures (RM) ANOVA Comparing Plaice Liver Body Burden Variables Among Areas and Between Years (2004, 2005)**

Variable	p values			
	Between Areas		Within Areas	
	Study versus Reference (SR)	Among References	Year	Year x SR
% moisture	0.724	0.538	0.237	0.708
% fat	0.861	0.380	0.486	0.694
Metals PC1	0.179	0.867	0.605	0.909
Arsenic	0.863	0.980	0.982	0.775
Cadmium	0.111	0.956	0.865	0.915
Copper	<b>0.024</b>	0.949	0.598	0.980
Iron	0.919	0.951	0.385	0.953
Manganese	0.690	0.559	0.633	0.735
Mercury	0.203	0.708	0.525	0.445
Selenium	0.935	<b>0.097</b>	0.280	0.915
Zinc	<b>0.001</b>	0.998	0.283	0.971
>C <sub>10</sub> -C <sub>21</sub> HCs	0.765	<b>0.041</b>	0.425	0.723
>C <sub>21</sub> -C <sub>32</sub> HCs	0.354	0.717	0.487	0.267

Note: - See Table 6-7 and Section 6.3.2.1 for further explanation of the RM ANOVA



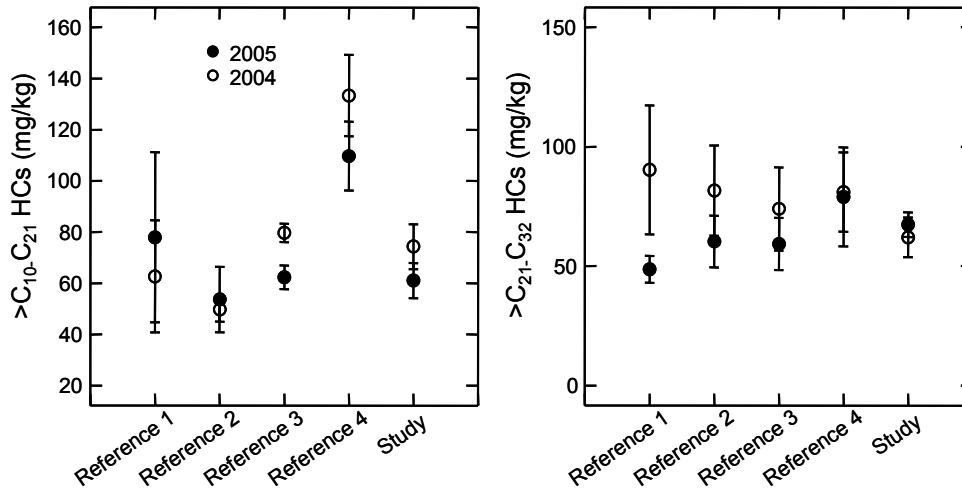
**Figure 6-11 Metals PC1 Scores, Copper and Zinc Concentrations for Plaice Liver Composites (2004, 2005)**

Note: - Values are Area means ± 1 SE (vertical bars); SE were based on variance among composites

More generally, except for selenium and >C<sub>10</sub>-C<sub>21</sub> HCs, there were no significant differences among Reference Areas across both years (Table 6-23). Many *p* values for the Between Areas Among References term were high (i.e., > 0.90, which suggests negative variance over time or carry-over effects) (Table 6-23). The Between Areas Among References (AR) term is tested against the Within Areas error term (Year x AR), or the variance of Reference Area differences between years. This error term was relatively large, because many Reference Area differences that occurred in 2004 were reversed in 2005. For example, copper and zinc concentrations were highest in Reference Area 4 in 2004 but lowest there in 2005 (Figure 6-11). The net effect would be convergence on “no Reference Area differences” over the long term (i.e., negative carry-over effects), although two years are too few to determine if this convergence is real. Note also that if the Year x AR interaction is relatively large, the Within Areas Year

and Year x SR terms, which are tested against Year x AR, are unlikely to be, and were not, significant (Table 6-23).

>C<sub>10</sub>-C<sub>21</sub> HCs were one of the few body burden variables to differ significantly and consistently among Reference Areas over the two years (Table 6-23). Concentrations were higher in Reference Area 4 than in other Areas in both years (Figure 6-12). A similar difference among Reference Areas was not evident for >C<sub>21</sub>-C<sub>32</sub> HCs. Instead, >C<sub>21</sub>-C<sub>32</sub> HCs provided an example of a strong Year x AR interaction, with the rank order of Reference Areas almost completely reversed from 2004 to 2005 (Figure 6-12).



**Figure 6-12 >C<sub>10</sub>-C<sub>21</sub> and >C<sub>21</sub>-C<sub>32</sub> HC Concentrations for Plaiice Liver Composites in 2004 and 2005**

Note: - Values are Area means ± 1 SE (vertical bars); SE were based on variance among composites

**Filletts**

Summary statistics for concentrations of detected substances are provided in Table 6-24. Raw data are provided in Appendix C-2. One fillet sample had HCs in the >C<sub>10</sub>-C<sub>21</sub> range, but the chromatogram for this sample did not indicate the presence of drill muds (Maxxam Analytics, pers. comm.).

**Table 6-24 Summary Statistics for Plaiice Fillet Body Burden (2004, 2005)**

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
>C <sub>10</sub> -C <sub>21</sub>	2005	Reference Area 1	3	3	<15	<15	<15			
		Reference Area 2	3	3	<15	<15	<15			
		Reference Area 3	3	3	<15	<15	<15			
		Reference Area 4	3	2	<15	16.0	<15			
		Reference Means								
		Study Area North	5	5	<15	<15	<15			
		Study Area South	5	5	<15	<15	<15			
		Study Means								
Arsenic	2004	Reference Area 1	3	0	1.90	2.90	2.60	2.47	0.51	21
		Reference Area 2	3	0	2.10	2.60	2.20	2.30	0.26	12
		Reference Area 3	3	0	2.60	3.50	3.30	3.13	0.47	15
		Reference Area 4	3	0	3.40	4.00	3.50	3.63	0.32	9
		Reference Means					2.90	2.88		
		Study Area	10	0	2.00	4.20	2.75	2.79	0.68	24
	2005	Reference Area 1	3	0	2.20	4.36	2.48	3.01	1.17	39



Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
Arsenic	2005	Reference Area 2	3	0	2.34	3.33	2.53	2.73	0.53	19
		Reference Area 3	3	0	2.99	4.27	3.88	3.71	0.66	18
		Reference Area 4	3	0	2.55	3.59	3.53	3.22	0.58	18
		Reference Means					3.11	3.17		
		Study Area North	5	0	2.25	3.09	2.97	2.84	0.34	12
		Study Area South	5	0	2.46	2.83	2.51	2.61	0.17	6
		Study Means				2.74	2.72			
Iron	2004	Reference Area 1	3	3	<15	<15	<15			
		Reference Area 2	3	3	<15	<15	<15			
		Reference Area 3	3	3	<15	<15	<15			
		Reference Area 4	3	3	<15	<15	<15			
		Reference Means								
		Study Area	10	9	<15	38.0	<15			
Mercury	2004	Reference Area 1	3	0	0.070	0.120	0.090	0.090	0.030	27
		Reference Area 2	3	0	0.070	0.100	0.090	0.090	0.020	18
		Reference Area 3	3	0	0.060	0.090	0.060	0.070	0.020	25
		Reference Area 4	3	0	0.050	0.080	0.080	0.070	0.020	25
		Reference Means					0.080	0.080		
		Study Area	10	0	0.040	0.100	0.090	0.080	0.020	25
	2005	Reference Area 1	3	0	0.060	0.080	0.070	0.070	0.010	14
		Reference Area 2	3	0	0.030	0.110	0.070	0.070	0.040	57
		Reference Area 3	3	0	0.070	0.140	0.110	0.110	0.040	33
		Reference Area 4	3	0	0.060	0.070	0.060	0.060	0.010	9
		Reference Means					0.080	0.080		
		Study Area North	5	0	0.040	0.100	0.080	0.070	0.020	32
		Study Area South	5	0	0.070	0.150	0.080	0.090	0.030	36
		Study Means					0.080	0.080		
Selenium	2004	Reference Area 1	3	3	<0.5	<0.5	<0.5			
		Reference Area 2	3	3	<0.5	<0.5	<0.5			
		Reference Area 3	3	3	<0.5	<0.5	<0.5			
		Reference Area 4	3	3	<0.5	<0.5	<0.5			
		Reference Means								
		Study Area	10	9	<0.5	0.50	<0.5			
	2005	Reference Area 1	3	3	<0.5	<0.5	<0.5			
		Reference Area 2	3	3	<0.5	<0.5	<0.5			
		Reference Area 3	3	2	<0.5	0.51	<0.5			
		Reference Area 4	3	3	<0.5	<0.5	<0.5			
		Reference Means								
		Study Area North	5	5	<0.5	<0.5	<0.5			
		Study Area South	5	5	<0.5	<0.5	<0.5			
		Study Means								
Strontium	2004	Reference Area 1	3	3	<1.5	<1.5	<1.5			
		Reference Area 2	3	3	<1.5	<1.5	<1.5			
		Reference Area 3	3	2	<1.5	1.5	<1.5			
		Reference Area 4	3	3	<1.5	<1.5	<1.5			
		Reference Means								
		Study Area	10	10	<1.5	<1.5	<1.5			
Zinc	2004	Reference Area 1	3	0	4.20	4.60	4.40	4.40	0.20	5
		Reference Area 2	3	0	4.30	4.80	4.30	4.47	0.29	6
		Reference Area 3	3	0	4.00	4.30	4.20	4.17	0.15	4
		Reference Area 4	3	0	3.80	4.00	3.90	3.90	0.10	3
		Reference Means					4.20	4.23		
		Study Area	10	0	3.40	4.80	4.20	4.20	0.36	8
	2005	Reference Area 1	3	0	3.80	4.60	4.20	4.20	0.40	10
		Reference Area 2	3	0	3.40	4.20	4.00	3.87	0.42	11
		Reference Area 3	3	0	4.60	4.70	4.70	4.67	0.06	1
		Reference Area 4	3	0	3.70	4.10	4.00	3.93	0.21	5
		Reference Means					4.23	4.17		
		Study Area North	5	0	3.80	4.60	4.20	4.16	0.36	9
		Study Area South	5	0	3.70	4.80	4.60	4.38	0.44	10
		Study Means					4.40	4.27		
% Fat	2004	Reference Area 1	3	0	1.10	1.90	1.70	1.57	0.42	27
		Reference Area 2	3	0	1.10	1.40	1.20	1.23	0.15	12
		Reference Area 3	3	0	2.20	3.60	2.50	2.77	0.74	27

Variable	Year	Area	n	n < EQL	Min	Max	Median	Mean	SD	CV %
% Fat	2004	Reference Area 4	3	0	1.10	3.10	2.20	2.13	1.00	47
		Reference Means					1.90	1.93		
		Study Area	10	0	1.00	3.30	1.95	1.99	0.67	33
	2005	Reference Area 1	3	0	1.30	1.80	1.70	1.60	0.26	17
		Reference Area 2	3	0	0.90	1.50	0.90	1.10	0.35	31
		Reference Area 3	3	0	1.50	2.60	2.00	2.03	0.55	27
		Reference Area 4	3	0	1.40	1.80	1.50	1.57	0.21	13
		Reference Means					1.53	1.58		
		Study Area North	5	0	0.80	1.70	1.50	1.32	0.40	30
		Study Area South	5	0	0.60	2.30	1.90	1.56	0.69	44
Study Means					1.70	1.44				
% Moisture	2004	Reference Area 1	3	0	77.00	80.00	78.00	78.33	1.53	2
		Reference Area 2	3	0	77.00	79.00	78.00	78.00	1.00	1
		Reference Area 3	3	0	77.00	81.00	79.00	79.00	2.00	3
		Reference Area 4	3	0	80.00	81.00	81.00	80.67	0.58	1
		Reference Means					79.00	79.00		
		Study Area	10	0	78.00	81.00	78.50	78.90	1.10	1
	2005	Reference Area 1	3	0	77.00	82.00	81.00	80.00	2.65	3
		Reference Area 2	3	0	80.00	83.00	83.00	82.00	1.73	2
		Reference Area 3	3	0	81.00	82.00	81.00	81.33	0.58	1
		Reference Area 4	3	0	79.00	81.00	81.00	80.33	1.15	1
		Reference Means					81.50	80.92		
		Study Area North	5	0	80.00	83.00	80.00	80.80	1.30	2
		Study Area South	5	0	78.00	82.00	80.00	79.80	1.79	2
Study Means					80.00	80.30				

Note: - All units are mg/kg except where indicated  
 - Statistics are reported to one more significant digit than what is given for EQL (see Table 6-4)

**Analyses of 2005 Data**

Moisture and fat content, and concentrations of metals, in plaice fillets did not differ significantly among Areas (Table 6-25). The lowest *p* values observed were for differences in arsenic concentrations between the Study versus Reference Areas, and for differences in zinc concentrations among the Reference Areas (Table 6-23). Arsenic concentrations in the Northern and Southern portion of the Study Area were lower than in the Reference Areas. Zinc concentrations in Reference Areas 2 and 4 were somewhat lower than in the other two Reference Areas (and in the Northern and Southern portions of the Study Area).

**Table 6-25 Results of Modified Nested ANOVA Comparing Plaice Fillet Body Burden Variables Among Areas (2005)**

Variable	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
% moisture	0.444	0.393	0.349	0.432	0.392
% fat	0.165	0.606	0.436	0.666	0.517
Arsenic	0.249	0.644	0.538	0.241	0.092
Mercury	0.255	0.422	0.274	0.778	0.712
Zinc	0.056	0.619	0.345	0.727	0.509

Notes: - MSE = variance among composites within Areas; MS(AR) = variance among Reference Areas  
 - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

Arsenic concentrations in fillets were uncorrelated with mercury and zinc concentrations (Table 6-26). Mercury and zinc concentrations were positively but not significantly correlated. Similar results were obtained in 2004 (Husky Energy 2005), suggesting that arsenic behaved differently than mercury and zinc, which behaved somewhat similarly.

**Table 6-26 Spearman Rank Correlations ( $r_s$ ) Among Plaice Fillet Burden Variables, and Between Those Variables and Composite Mean Gutted Weights (2005)**

	% moisture	% fat	Arsenic	Mercury	Zinc
Gutted weight	-0.132	<b>0.534*</b>	-0.004	0.670**	<b>0.536**</b>
% moisture		-0.075	<b>0.431*</b>	-0.096	-0.285
% fat			-0.015	0.256	0.388
Arsenic				-0.165	-0.036
Mercury					0.328

Notes: -  $n = 22$  composites  
 -  $*p \leq 0.05$ ;  $**p \leq 0.01$ ;  $r_s$  at  $p \leq 0.05$  in bold

Fat content and concentrations of zinc and mercury increased significantly with increasing size (i.e., composite mean gutted weights) (Table 6-26). For all three body burden variables, the positive correlations were a function of the large size differences between Reference Area 3 fish, plus the much smaller size differences within and among the other five Areas. With Reference Area 3 excluded, correlations with size were not markedly reduced, and were still significant at  $p \leq 0.10$  for fat content ( $r_s = 0.454$ ) and  $p \leq 0.01$  for mercury concentrations ( $r_s = 0.644$ ). The positive correlation between fat content and size may indicate that larger fish store more fat in fillets/muscle versus liver than smaller fish. As was the case for crab claws and plaice liver, positive correlations between size and metal concentrations may be evidence of biomagnification or changes in physiology and uptake/elimination rates with size.

Moisture content was significantly positively correlated with arsenic concentrations, but not mercury and zinc concentrations. These correlations suggest that wet rather than dry weight arsenic concentrations may have been more similar among samples (or given the absence of any differences among Areas, may have varied more among Areas).

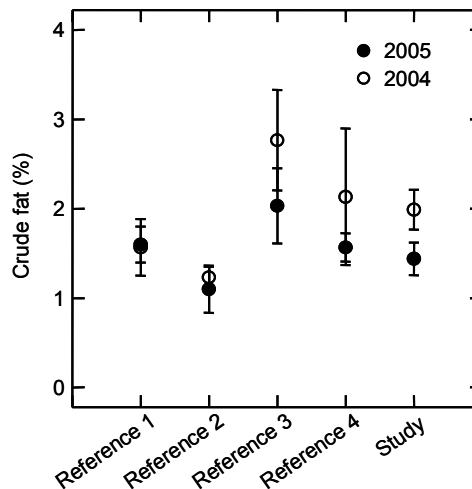
**Comparison of 2005 versus 2004 Results**

Except for fat content, terms in RM ANOVA comparing plaice fillet body burden variables among Areas and between years (2004, 2005) were not significant even at  $p \leq 0.10$  (Table 6-27). To some extent, differences among Reference Areas tended to reverse from 2004 to 2005, as was the case for liver (see above; the changes in Reference Area differences are the error variance for three of the four terms in the ANOVA). However, differences over both space and time were also small, and the  $F$  statistics and associated  $p$  values for both within- and between-year tests in this report and Husky Energy (2005) approximate random distributions. Fat content differed mostly among Reference Areas in both years, with values greater in Reference Areas 3 and 4 (Figure 6-13).

**Table 6-27 Results of Repeated Measures ANOVA Comparing Plaice Fillet Body Burden Variables Among Areas and Between Years (2004, 2005)**

Variable	p values			
	Between Areas		Within Areas	
	Study versus Reference (SR)	Among References	Year	Year × SR
% moisture	0.612	0.764	0.197	0.813
% fat	0.956	<b>0.058</b>	0.111	0.653
Arsenic	0.642	0.142	0.701	0.549
Mercury	0.736	0.733	0.915	0.964
Zinc	0.893	0.543	0.995	0.807

Note: - See Table 6-7 and Section 6.3.2.1 for further explanation of the RM ANOVA



**Figure 6-13 Crude Fat Content for Plaice Fillet Composites (2004 and 2005)**

Note: - Values are Area means ± 1 SE (vertical bars); SE were based on variance among composites

### 6.4.3 Taste Tests

No significant difference in taste was noted between crab from the Study and Reference Areas in both the triangle and hedonic scaling tests. Panelists for the triangle test were successful in discriminating only 8 out of 24 samples. These results were not significant at  $\alpha = 0.05$  (Appendix C-4). ANOVA statistics for hedonic scaling are provided in Table 6-28. The results were not significant ( $p = 0.67$ ;  $\alpha = 0.05$ ), and from the frequency histogram (Figure 6-14), samples from both the Study and Reference Areas were assessed similarly for preference. From ancillary comments (Table 6-29 and 6-30, and Appendix C-4), there were no consistent comments identifying abnormal or foreign odour or taste.

**Table 6-28 Analysis of Variance for Preference Evaluation by Hedonic Scaling of Crab (2005)**

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.333333	1	0.333333	0.180569	0.672866	4.051742
Within Groups	84.91667	46	1.846014			
Total	85.25	47				

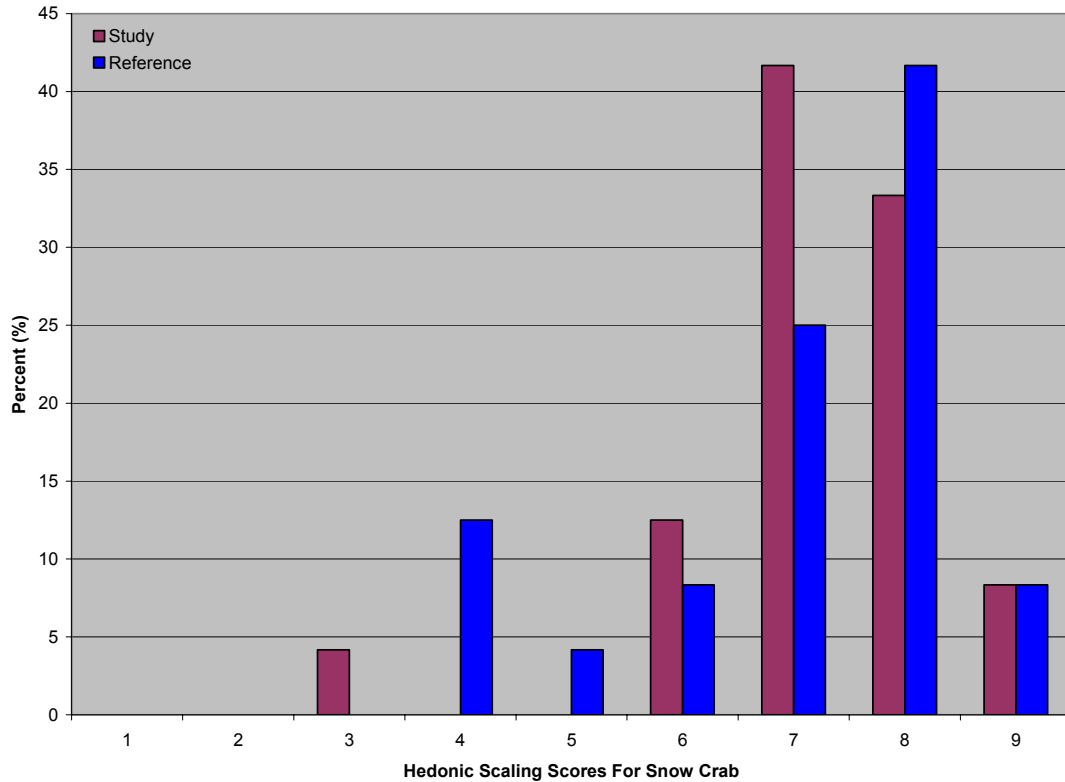


Figure 6-14 Crab Frequency Histogram for Hedonic Scaling Sensory Evaluation (2005)

Table 6-29 Summary of Comments from the Triangle Test for Crab (2005)

Reference Area (RA) Correctly Identified as Odd Sample	Study Area (SA) Correctly Identified as Odd Sample
To be honest I could not detect any noticeable difference. 294 (RA) may have been a little bitter.	Liked the odd sample better. 331(RA) and 653 (RA) were strong in taste.
	Sample 581(SA) tasted more salty.
	Very close, very little difference detectable on odour and or flavour.
Reference Area Incorrectly Identified as Odd Sample	Study Area Incorrectly Identified as Odd Sample
331(RA) had a sweeter taste.	539 (SA) tasted a little sweeter.
Much the same. Sample 331 (RA) didn't seem as strong.	Hard to distinguish.
Samples 331(RA) and 581(SA) tasted slightly sweeter than the last sample. These two samples also had more of a sea fresh taste and odour.	Couldn't detect very much difference between samples.
I couldn't tell a difference in the odour, but 954 (SA) and 532 (RA) tasted more flavourful than 252 (RA).	Less sweet taste was noted in samples 294 (RA) and 104 (SA).
Tastes like real crab.	294 (RA) and 104 (SA) seemed to have a stronger taste.
252 (RA) had a bland taste.	No difference in odour. 216 (SA) tasted a little more bland than the other two.
Found 532 (RA) to be different in flavour and odour.	

**Table 6-30 Summary of Comments from Hedonic Scaling Tests for Crab (2005)**

<b>Prefer Reference Area (RA)</b>	<b>Prefer Study Area (SA)</b>
No significant difference to me. Actually they were both pretty tasty.	277 (SA) tasted a little sweeter.
Both tasted pretty much the same to me.	277 (SA) has a much better flavour (more tangy).
344 (RA) natural, characteristic odour present. 577 (SA) neutral odour. 344 (RA) flavour (good, normal). 577 (SA) flavour OK, but less desirable than 344 (RA). No off flavour or odours detected.	No significant difference to me. Actually they were both pretty tasty.
Prefer 344 (RA) for odour and flavour. 577 (SA) too salty.	Both tasted pretty much the same to me.
Not a significant difference. 344 (RA) slightly more flavourful.	213 (SA) had a sweeter, less fishy flavour. 878 (RA) had bits of shell and cartilage, making it less desirable. 878 (RA) also had more of a fishy after taste.
344 (RA) had a more robust flavour. Both had a pleasant odour.	The taste of 213 (SA) is preferred. 213 (SA) also had a slightly preferred odour.
Good taste, chalky, tough texture - 213 (SA). Sweet taste, slightly chalk texture - 878 (RA).	213 (SA) and 878 (RA) has a very nice odour. 878 (RA) has a more salty taste.
I couldn't tell any difference in the two samples.	Not a lot of difference between the two.
526 (SA) I found had more of a sharp after taste.	I couldn't tell any difference in the two samples.
Found both very similar. Better texture with 534 (RA).	Very normal flavour on both samples.
Very normal flavour on both samples.	Taste and smell of 526 (SA) was very pleasing. 534 (RA) taste was good, but odour was a bit off. Nothing severe, but not as good as 526 (SA).
No real difference between the samples.	No real difference between the samples.

For plaice, panelists for the triangle test were successful in discriminating 10 out of 24 samples. These results are not significant at  $\alpha = 0.05$  (Appendix C-4). ANOVA statistics for hedonic scaling are provided in 6-31. These results were significant ( $p = 0.05$ ;  $\alpha = 0.05$ ) with a preference for samples from the Reference Areas (Figure 6-15). However, from ancillary comments (Table 6-32 and 6-33; Appendix C-4), there were no consistent comments identifying abnormal or foreign odour or taste for either the triangle or hedonic test. Two panelists felt the Study Area sample had an “off” or rancid taste. Two panelists felt the Reference Area sample had an “off” or sour taste. Most comments focused on texture with a majority of panelists preferring the texture of Reference Area samples.

**Table 6-31 Analysis of Variance Preference Evaluation by Hedonic Scaling of Plaice (2004)**

<b>Source of Variation</b>	<b>SS</b>	<b>df</b>	<b>MS</b>	<b>F</b>	<b>P-value</b>	<b>F crit</b>
Between Groups	8.333333	1	8.333333	4.096171	0.048811	4.051742
Within Groups	93.58333	46	2.03442			
Total	101.9167	47				

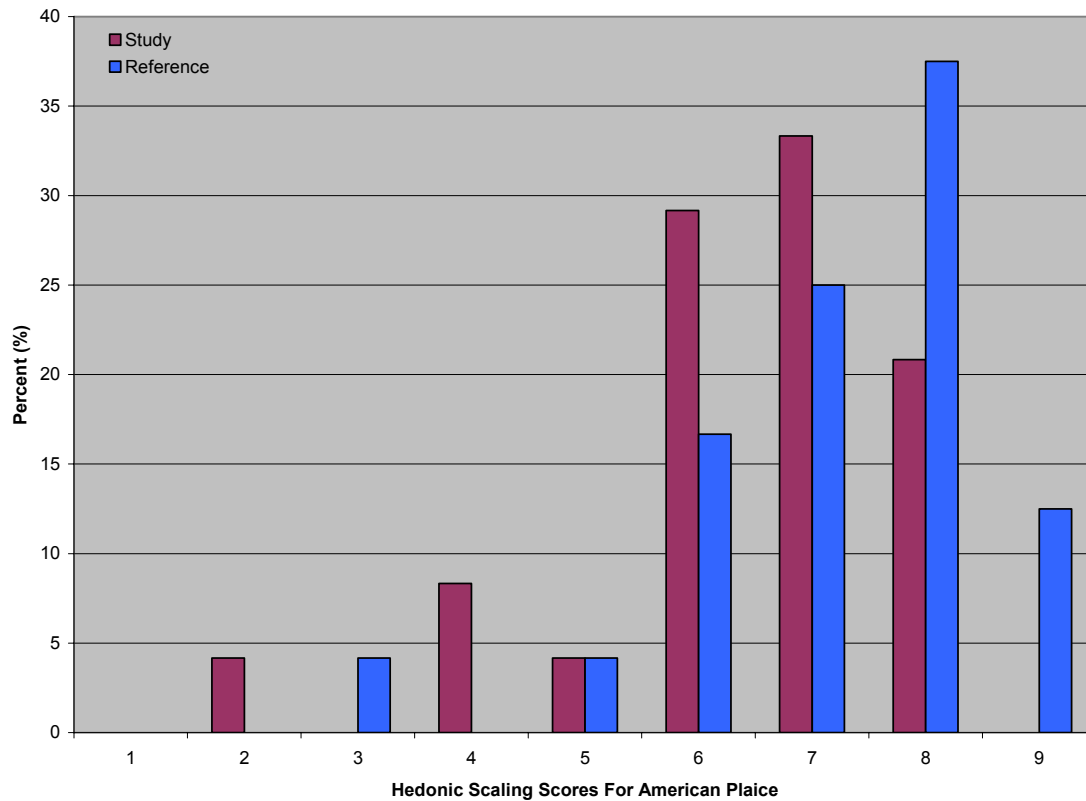


Figure 6-15 Paice Frequency Histogram for Hedonic Scaling Sensory Evaluation (2005)

Table 6-32 Summary of Comments from the Triangle Test for Plaice (2005)

Reference Area (RA) Correctly Identified as Odd Sample	Study Area (SA) Correctly Identified as Odd Sample
647 (RA) had a much stronger flavour. Texture more firm and less sweet.	All three samples were very similar in taste and texture. Sample 980 (SA) had a plain taste. The other samples tasted more fishy.
	Odd sample had more acceptable flavour and odour.
	All samples very similar.
	223 (SA) was a tougher and grittier taste.
	223 (SA) was a little more grainy in terms of texture.
	039 (RA) and 426 (RA) had better taste and texture.
	Very difficult to tell a difference.
Reference Area Incorrectly Identified as Odd Sample	Reference Area Incorrectly Identified as Odd Sample
Sample 813 (RA) was most preferred.	832 (SA) had a firmer consistency. Other two were too watery.
813 (RA) is milder.	815 (SA) bland and mushy
No difference in taste. 426 (RA) has a different texture.	Little more subtle flavour than other two.
Sample 039 ((RA) is firmer in texture and also more tasty.	Off taste in 832 (SA), maybe rancidity.
	832 (SA) tastes blander to me and the texture is not as grainy as the other two.
	No significant difference to me. 648 (SA) may have been grainier.
	Not very much difference between all three.

**Table 6-33 Summary of Comments from the Hedonic Scaling Test for Plaice (2005)**

Prefer Reference Area (RA)	Prefer Study Area (SA)
619 (SA) had a strong fishy taste after a couple of seconds. 835 (RA) tasted somewhat the same, little less fishy taste.	835 (RA) seemed milder and somewhat bland. Also 835 (RA) had whiter appearance. 619 (SA) more flavourful. Both had same consistency, chewy, slightly dry.
Stronger (undesirable to me) flavour on sample 619 (SA).	447 (SA) had more flavour.
Sample 698 (RA) has a stronger taste.	Samples were very similar in taste texture and odour.
Sample 698 (RA) had a much better aroma and mouth feel.	Slightly off taste on 874 (RA).
Samples were very similar in taste texture and odour.	252 (SA) bit grainy and somewhat sweet. 874 (RA) much the same as 252 (SA), bit less grainy and liked texture better than 252 (SA).
447 (SA) had a very fishy taste. 698 (RA) was very juicy and tasty.	Couldn't taste any difference between samples.
Sample 698 (RA) was good-not as fishy.	781 (RA) had a stronger taste, somewhat sour.
The taste and texture of both samples are very similar.	
Taste is very similar. 874 (RA) has a better appearance and texture.	
252 (SA) bit grainy and somewhat sweet. 874 (RA) much the same as 252 (SA), bit less grainy and liked texture better than 252 (SA).	
The texture is really poor on the 252 (SA) sample and initially there is an off taste.	
874 (RA) nicer texture, 252 (SA) grainy. Overall taste, not much difference.	
Couldn't taste any difference between samples.	
781 (RA) had nicer and firmer consistency. 038 (SA) was too wet and mushy.	
038 (SA) was drier and seemed like it was in pieces that had been packed together. 781 (RA) was moister.	
781 (RA) had better consistency and the flavour of the fish was more pronounced	

#### 6.4.4 Fish Health Indicators

A total of 180 plaice were examined for early warning effects on fish health. Sixty (60) fish were sampled in the Study Area, with 29 fish taken in the Northern portion and 31 fish in the Southern portion. Thirty (30) fish were also sampled from each of four Reference Areas. The full report on plaice health indicators is provided in Appendix C-3. Highlights of results are provided below.



**6.4.4.1 Mixed Function Oxygenase**

MFO enzyme activities, measured as EROD, in mature females, immature females, and males (matures and immatures pooled) from the various Areas are summarized in Figures 6-16 and 6-17. Area medians for mature females were approximately 20 pmol/min/mg protein. MFO activities for immature females and males were higher (all Area medians were greater than 20 pmol/min/mg protein) and more variable within Areas. The complete data set on fish from the 2005 survey is provided in Appendix C-3 (Annex D).

Results were compared among Areas in modified nested ANOVA (Table 6-34). There were no significant differences among Areas for any of the three groups. MFO activities for mature females were log-transformed for analysis to reduce the influence of the one high value in the Northern portion of the Study Area (Figure 6-16A).

**Table 6-34 Results of Nested ANOVA Comparing MFO Activities in Plaice (2005)**

Group	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
<b>Mature females</b> <sup>a</sup>	0.516	0.668	<i>0.680</i>	0.737	<i>0.746</i>
<b>Immature females</b>	0.119	0.619	<i>0.389</i>	0.750	<i>0.588</i>
<b>Males</b>	0.174	0.611	<i>0.420</i>	0.993	<i>0.989</i>

- Notes:
- See Appendix C-3 (Annex B) for details on application and interpretation of modified nested ANOVA
  - MSE = variance among fish within Areas; MS(AR) = variance among Reference Areas
  - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)
  - <sup>a</sup> MFO activities were log-transformed

**6.4.4.2 Gross Pathology**

Two fish, one from Reference Area 1 and one from Reference Area 4, displayed gill achromasia or white gill (Appendix C-3, Annex H, Photo 1), whereas another fish from Reference Area 1 exhibited a skin lesion.

**6.4.4.3 Haematology**

Blood smears were examined for various types of cells. The red blood cells of all fish appeared to be normal in size and shape. Coloration was also similar indicating a similar degree of haemoglobinization.

A differential cell count of lymphocytes, neutrophils and thrombocytes was carried out on a total of 170 fish. It was not possible to withdraw blood from five fish and blood smears of five other fish were not suitable for cell counting due to clotting problems. For the other blood smears, 200 cells were counted per fish and the results were expressed as mean percentage ± standard deviation of each cell type (6-35).

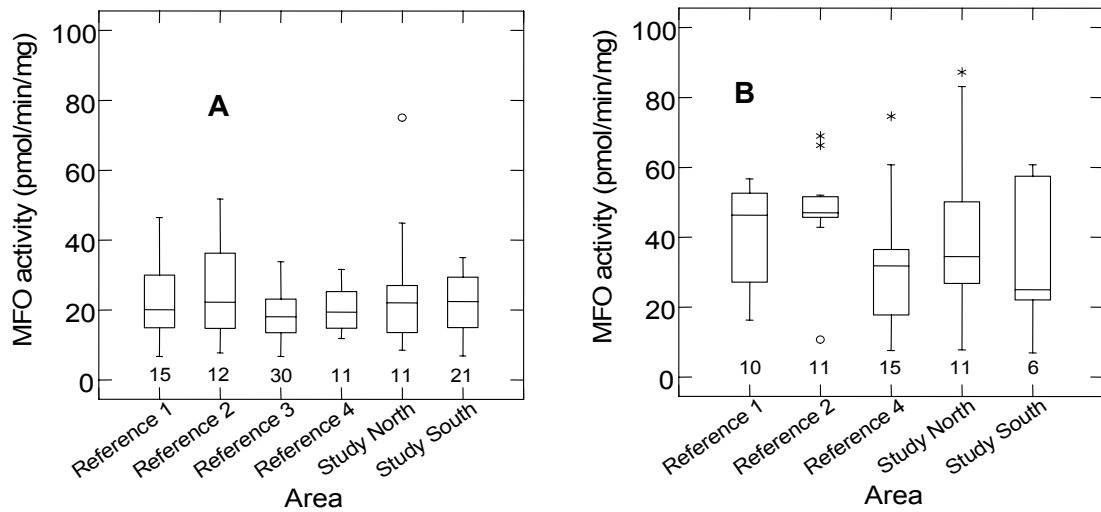


Figure 6-16 MFO Activity in (A) and (B) Immature Females (2005)

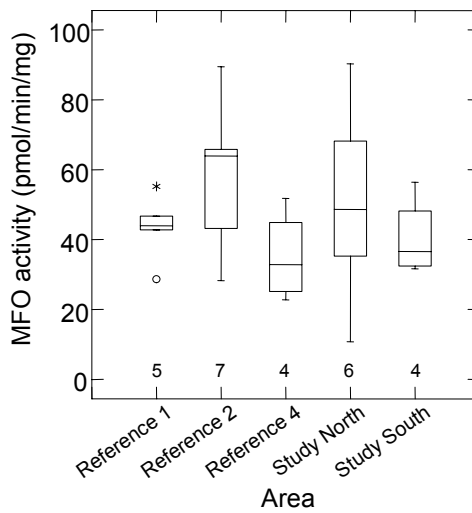


Figure 6-17 MFO Activity in Males (2005) (All Maturity Stages Pooled)

- Notes:
- Horizontal line in middle of box = median
  - Box = 25<sup>th</sup> to 75<sup>th</sup> percentile
  - Vertical lines = whiskers; include all values within 1.5 Hspread (75<sup>th</sup> minus 25<sup>th</sup> percentiles)  
The box whiskers will often include all the points, especially when n is small
  - \* Asterisks are outside values, >1.5 Hspreads from the 25<sup>th</sup> or 75<sup>th</sup> percentiles
  - ° Circles are far outside values, >3 Hspreads from the 25<sup>th</sup> or 75<sup>th</sup> percentiles
  - The number under each box is the sample size

**Table 6-35 Frequencies of Blood Cell Types in Plaice (2005)**

Area	No. fish	% lymphocytes	% thrombocytes	% neutrophils
Reference 1	28	70.3 ± 3.5	29.1 ± 3.6	0.61 ± 0.69
Reference 2	28	70.4 ± 4.7	29.4 ± 4.8	0.18 ± 0.39
Reference 3	29	69.3 ± 6.3	30.4 ± 6.4	0.10 ± 0.41
Reference 4	27	70.1 ± 6.0	29.3 ± 5.9	0.59 ± 0.84
All References	112	70.0	29.6	0.37
North Study	29	62.9 ± 8.8	35.8 ± 9.0	1.24 ± 0.99
South Study	29	70.2 ± 5.6	29.7 ± 5.6	0.14 ± 0.35
Both Study	58	66.6	32.7	0.69

Notes: - All data are means ± standard deviations  
 - All References = means of the three Reference Area means; Both Study = means of the North and South Study Area means

The complete data set on the different cells examined is provided in Appendix C-3 (Annex E) and a representative photograph of a blood smear (Photo 2) is included in Appendix C-3 (Annex H).

Percentages of lymphocytes and thrombocytes were compared among Areas using modified nested ANOVA. For all tests, *p* values for the two variables were similar (Table 6-36), because the two percentages summed to almost 100% for all fish (i.e., the two variables were almost perfectly negatively correlated). Percentages of the two cell types did not differ significantly among Reference Areas (Table 6-36), and all Reference Area means were approximately 70% lymphocytes and 30% thrombocytes (Table 6-36). Differences between the Northern and Southern portion of the Study Area were significant. Blood samples from fish from the Northern portion of the Study Area had fewer lymphocytes and more thrombocytes than blood samples from fish from the Southern portion (Table 6-36). The Study versus Reference contrast was also significant, but only because of the difference between the Northern portion of the Study Area and all other Areas. Percentages of lymphocytes and thrombocytes for fish from the Southern portion of the Study Area were similar to those for Reference fish (i.e., 70% lymphocytes; 30% thrombocytes; Table 6-35).

**Table 6-36 Results of Nested ANOVA Comparing Percentages of Blood Cell Types in Plaice (2005)**

Group	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
% lymphocytes	0.910	0.002	<0.001	0.004	0.001
% thrombocytes	0.861	0.005	<0.001	0.008	0.002

Notes: - See Appendix C-3 (Annex B) for details on application and interpretation of modified nested ANOVA  
 - MSE = variance among fish within Areas; MS(AR) = variance among Reference Areas  
 - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when *p* ≥ 0.25 for Among References contrast)

**6.4.4.4 Histopathology**

**Liver Histopathology**

Results of the detailed histopathological studies carried out on liver tissues of plaice from the Reference and Study Areas are summarized in Table 6-37. The complete data set is provided in Appendix C-3 (Annex F).

Sixty (60) fish from the Study Area and 120 fish from the Reference Areas were examined and no cases of megalocytic hepatitis, foci of cellular alteration (including basophilic foci, clear cell foci and eosinophilic foci), carcinoma, cholangioma, cholangiofibrosis or hydropic vacuolation were observed.

**Table 6-37 Number of Plaice with Specific Types of Hepatic Lesions and Prevalence of Lesions (2005)**

Variable	Area								
	Ref 1	Ref 2	Ref 3	Ref 4	All References	North Study	South Study	Both Study	
No. fish	30	30	30	30	120	29	31	60	
Nuclear pleomorphism	No.	1	0	0	0	1	0	0	0
	%	3.3	0.0	0.0	0.0	0.8	0.0	0.0	0.0
Megaolocytic hepatitis	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Basophilic foci	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Clear cell foci	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Eosinophilic foci	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Macrophage aggregation <sup>a</sup>	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Hepatocellular carcinoma	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cholangioma	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Cholangio-Fibrosis	No.	0	0	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Inflammatory response	No.	1	0	1	0	2	0	1	1
	%	3.3	0.0	3.3	0.0	1.7	0.0	3.2	1.7
Hepatocellular vacuolation	No.	1	0	5	0	6	0	3	3
	%	3.3	0.0	16.7	0.0	5.0	0.0	9.7	5.0
Biliary parasites	No.	2	1	2	2	7	3	4	7
	%	6.7	3.3	6.7	6.7	5.8	10.3	12.9	11.7

Notes: - <sup>a</sup> Moderate to high aggregation (> 3 on a 0 to 7 relative scale)

Nuclear pleomorphism (Appendix C-3, Annex H, Photo 4) occurred in one fish from Reference Area 1.

The frequencies of macrophage aggregates in livers of fish from the various Areas were low (0 to 3 rating on a relative scale of 0 to 7) and no cases of moderate to high aggregation (4 or higher on the relative scale) were observed.

One fish from the Southern portion of the Study Area, one fish from Reference Area 1 and another from Reference Area 3 exhibited an inflammatory response (Appendix C-3, Annex H, Photo 4).

Six fish from the Reference Areas (5%) and three fish from the Study Area (5%) displayed a “patchy distribution” of hepatocellular vacuolation. This type of vacuolation is likely a reflection of gonadal maturational stage.

An infestation of the biliary system with a myxosporean parasite (Appendix C-3, Annex H, Photo 5), possibly *Myxidium sp.*, was observed in 5.8% of fish from the Reference Areas and in 11.7% of fish from the Northern and Southern portion of the Study Area.

Fisher’s Exact Test was used to compare presence versus absence of biliary parasites between the Study and Reference Areas (= SR contrast). All fish were pooled for the analysis, since there was no reason to expect differences between sexes or maturity stages. Incidences of parasites were low, so only the SR contrast was tested. Other liver abnormalities were rare or absent and were not statistically analyzed.

Incidences of biliary parasites did not differ significantly between the Northern and Southern portion of the Study Area and the pooled Reference Areas (Fisher’s Exact Test  $p = 0.24$ ).

The observations on parasitism are of general interest but the absence or very low incidence of liver lesions that have been associated with chemical toxicity are more relevant from an EEM perspective.

**Gill Histopathology**

One gill sample from Reference Area 1 and one from Reference Area 4 were missing. All other samples were processed in the same manner, however one sample from Reference Area 1 and one from Reference Area 3 could not be accurately read, possibly due to mechanical damage during sample collection. Also, three fish from the Reference Areas (one from Reference Area 1 and two from Reference Area 4), displayed extensive proliferation of ovoid and pale staining cells, or X-cells, in the interlamellar spaces of secondary lamellae (Appendix C-3, Annex H, Photo 6) and tissue structure was altered to such an extent that it was not possible to count the secondary lamellae in these samples.

Detailed histopathological studies were thus carried out on gill tissues of 113 fish from the four Reference Areas and 60 fish from the Northern and Southern portion of the Study Area (Table 6-38). The complete data set on fish from the 2005 survey is provided in Appendix C-3 (Annex G).

**Table 6-38 Occurrence of Different Stages and Oedema Condition in Plaice Gill (2005)**

Variable	Area							
	Ref 1	Ref 2	Ref 3	Ref 4	All References	North Study	South Study	Both Study
Number of fish	27	30	29	27	113	29	31	60
Stage 1 <sup>a</sup> : Thin lamellae	36.2 ± 17.8	40.5 ± 18.1	31.7 ± 14.6	41.0 ± 14.1	37.4	44.6 ± 18.3	37.9 ± 15.5	41.3
Stage 2 <sup>a</sup> : Distal hyperplasia	45.9 ± 18.3	40.7 ± 15.9	43.2 ± 17.1	43.8 ± 12.2	43.4	39.4 ± 17.3	36.0 ± 15.1	37.7
Stage 3 <sup>a</sup> : Epithelial lifting	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Stage 4a <sup>a</sup> : Tip hyperplasia	17.8 ± 15.6	18.8 ± 14.2	25.2 ± 14.7	15.2 ± 10.6	19.2	15.8 ± 14.4	26.2 ± 19.2	21.0

Variable	Area							
	Ref 1	Ref 2	Ref 3	Ref 4	All References	North Study	South Study	Both Study
Stage 4b <sup>a</sup> : Telangiectasis	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Stage 5 <sup>a</sup> : Basal hyperplasia	56.5 ± 33.8	40.3 ± 22.1	58.2 ± 25.1	39.3 ± 21.5	48.6	45.7 ± 21.4	47.9 ± 28.4	46.8
Stage 6 <sup>a</sup> : Fusion	0.07 ± 0.35	0.00	0.00	0.00	0.02	0.12 ± 0.44	0.00	0.06
Oedema condition <sup>b</sup>	1.33 ± 0.71	1.24 ± 0.61	1.06 ± 0.45	0.93 ± 0.31	1.14	1.12 ± 0.65	1.35 ± 0.55	1.24

- Notes:
- All data are means ± standard deviations
  - <sup>a</sup> Mean percentage of lamellae presenting the stage
  - <sup>b</sup> Mean of rating on a relative 0 to 3 scale
  - All References = means of the four Reference Area means; Both Study = means of the North and South Study Area means

Epithelial layers of secondary lamellae may vary in thickness. All the fish studied displayed a variable percentage of thin lamellae or Stage 1 (Appendix C-3, Annex H, Photo 7) and most of them also displayed a variable percentage of lamellae showing an increase in the number of epithelial layers occurring at the lamellar tips (tip hyperplasia or Stage 4a; Appendix C-3, Annex H, Photo 8), at the lamellar bases (basal hyperplasia or Stage 5; Appendix C-3, Annex H, Photo 9) or across the full length of the lamellae (distal hyperplasia or Stage 2; Appendix C-3, Annex H, Photo 10).

Statistical comparisons were carried out with modified nested ANOVA on percentages of thin lamellae and hyperplasia (tip, basal and distal) as well as on degree of oedema (Table 6-39).

**Table 6-39 Nested ANOVA Comparing Some Gill Histopathology Variables in Plaiçe (2005)**

Group	<i>p</i> values				
	Among References	Between Study		Study versus Reference	
	Error=MSE	Error=MS(AR)	Error=MSE	Error=MS(AR)	Error=MSE
Stage 1	0.108	<i>0.421</i>	0.185	<i>0.447</i>	0.213
Stage 2	0.782	0.989	<i>0.993</i>	0.042	<i>0.042</i>
Stage 4a	0.063	<i>0.232</i>	0.020	<i>0.851</i>	0.748
Stage 5	0.009	<i>0.989</i>	0.977	<i>0.872</i>	0.726
Oedema condition	0.113	<i>0.193</i>	0.019	<i>0.351</i>	0.119

- Note:
- See Appendix C-3 (Annex B) for details on application and interpretation of modified nested ANOVA
  - MSE = variance among fish within Areas; MS(AR) = variance among Reference Areas
  - *Italics* indicate *p* values that should be used for tests of the Between Study and Study versus Reference contrasts, following recommendations in Quinn and Keough (2002; i.e., test against MSE only when  $p \geq 0.25$  for Among References contrast)

Distal hyperplasia was similar among Reference Areas and between the Northern and Southern portion of the Study Area, but was significantly different between Study and Reference Areas. However, the degree of hyperplasia was lower in fish from the Study Area. Significant differences were also observed among Reference Areas for basal hyperplasia and between the Northern and Southern portion of the Study Area for tip

hyperplasia. Altogether, the results on hyperplasia in plaice indicate that a degree of variability in lamellar thickness is a natural occurrence.

With respect to microstructural changes which have been associated with chemical toxicity and could be more pathological in nature and thus of special note in EEM programs, these were absent or rarely observed (Table 6-38). Fusion (Appendix C-3, Annex H, Photo 11) was seen in only three fish, two collected in the Northern portion of the Study Area and one collected in Reference Area 1. No cases of epithelial lifting or telangiectasis were observed in any Area. The levels of oedema (rated on a 0 to 3 relative scale) were quite low in all Areas.

## **6.5 Summary of Findings**

### **6.5.1 Biological Characteristics**

#### **6.5.1.1 Crab**

Crab size and frequencies of recent moult for the 272 crab used in body burden analyses in 2005 did not differ significantly among Reference Areas, between the Northern and Southern portion of the Study Area, or between the Study and Reference Areas. Frequency of recent moults for the Northern and Southern portions of the Study area and all Reference Areas pooled was approximately 50%.

Carapace width of crab used in body burden analyses in 2005 was approximately 30% less than carapace width of crab used in 2004.

In both 2004 and 2005, smaller crab were more likely to be recent moults.

#### **6.5.1.2 Plaice**

Plaice liver and body burden composites usually consisted of a mix of larger mature females, and smaller males and immature females. Therefore, size varied considerably and was not normally distributed within composites.

Mean gutted weight for the three plaice composites collected from Reference Area 3 in 2005 were approximately double mean weights in composites from other Areas, because all Reference Area 3 fish were large mature females. In contrast, differences in composite mean weights in 2004 were smaller and not significant.

### **6.5.2 Body Burden**

#### **6.5.2.1 Crab**

HCs were not detected in crab claw samples in 2004 and 2005.

Concentrations of arsenic, copper, mercury, selenium, silver, and zinc in crab claws were positively correlated over all 2004 and 2005 samples (i.e., higher concentrations of these metals co-occurred). Boron concentrations were uncorrelated, and strontium concentrations were negatively correlated, with concentrations of the six correlated metals. Other metals were rarely or never detected.

Moisture and fat content did not differ significantly among Areas in 2005. Concentrations of most metals were similar between the Reference Areas and the Southern portion of the Study Area. Concentrations were higher in the Northern portion of the Study Area.

In 2005, crab size was negatively correlated with fat content and positively correlated with concentrations of most metals (strontium was a notable exception). Frequencies of recent moult were uncorrelated with body burden variables.

There were no consistent and significant differences in moisture and fat content, and metal concentrations, between the Study and Reference Areas over both EEM years (2004 and 2005). There were also no significant changes in Study and Reference Area differences between the two EEM years, probably because differences were negligible in both years.

### 6.5.2.2 Plaice

#### Liver

$>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HCs were detected in every plaice liver composite in 2005; as they were in every liver composite in 2004, except in one composite with very high EQL. These HCs did not resemble drill muds. Peaks observed on chromatograms were consistent with those expected for extracted fatty acid compounds.

Arsenic, cadmium, copper, iron, manganese, mercury, selenium and zinc were detected in every 2004 and 2005 liver composite. Other metals were rarely or never detected in liver samples in either year.

Based on multivariate analyses of all liver samples from 2004 and 2005, iron and manganese concentrations were largely uncorrelated with the other six frequently detected metals. In other words, higher concentrations of arsenic, cadmium, copper, mercury, selenium and zinc tended to co-occur, but iron and manganese “behaved differently”.

In 2005, moisture content, fat content, metal concentrations and HC concentrations either did not differ significantly among Areas, or differed mostly among Reference Areas or between the Northern portion and the Southern portion of the Study Area. Overall, Study Area metal and HC concentrations were generally within or below the Reference Area range.

Liver moisture and fat content were strongly negatively correlated in both 2005 and 2004 samples. In 2005, plaice body size (i.e., composite mean gutted weights) was significantly positively correlated with concentrations of most metals. The positive correlation was a function of both the large size differences between Reference Area 3 and other Areas, and the much smaller size differences between and within the other Areas. In both 2004 and 2005,  $>C_{10}-C_{21}$  and  $>C_{21}-C_{32}$  HC concentrations were positively correlated with fat content. As noted above, the HCs detected appear to be fatty acids, which can be included in both HC and fat content measurements. The relationship between HCs and fat might therefore at least be partly analytical.

There were few consistent differences in liver body burden variables between the Study and Reference Areas over both EEM years (2004 and 2005). Copper and zinc were minor exceptions, with concentrations significantly lower in the Study Area mostly



because other sources of variance were minimal. Study versus Reference differences also did not change significantly from 2004 to 2005 for any variable, probably because differences in both years were small. Overall, the major source of variance was probably the tendency for differences among Reference Areas to reverse from 2004 to 2005.

### **Filletts**

HCs and most metals were rarely or never detected in plaice fillet samples in either 2004 or 2005. Arsenic, mercury and zinc were detected in every fillet sample in both years.

In 2005, moisture and fat content, and concentrations of arsenic, mercury and zinc, did not differ significantly among Areas.

As was the case for crab claw and plaice liver, concentrations of some metals (i.e., mercury and zinc for fillets) increased with increasing body size in 2005 samples. Fat content of fillet samples also increased with size. Like liver, these correlations were as much or more a function of the small size differences among and within Areas other than Reference Area 3 versus the large size difference between that and other Areas.

In comparisons of both EEM years (2004 and 2005), moisture content and metal concentrations in fillets did not differ significantly over either space or time. Fat content was higher in Reference Areas 3 and 4 in both years.

### **6.5.3 Taste Tests**

There was no difference in taste between Study and Reference Area crab.

Panellists did not distinguish between Study and Reference Area plaice in the triangle test, but did show a preference for Reference Area plaice in the hedonic scaling test. However, there were no consistent comments resulting from the plaice tests that identified abnormal or foreign odour or taste which would normally be associated with taint. Most comments from panellist focussed on texture with a majority of panellists preferring the texture of Reference Area samples.

### **6.5.4 Fish Health Indicators**

There were no significant differences in MFO enzyme activities in mature females, immature females or males among Reference Areas, between the Northern and Southern portion of the Study Area, or between the Study and Reference Areas.

With respect to gross pathology, no external or internal lesions were noted except for one fish from Reference Area 1 and one from Reference Area 4 which displayed gill achromasia, and one fish from Reference Area 1 which had a skin lesion.

Percentages of blood lymphocytes and thrombocytes were similar in fish from all the Reference Areas as well as from the Southern portion of the Study Area. However, blood samples from the Northern portion of the Study Area had fewer lymphocytes and more thrombocytes than the other blood samples.

Regarding liver histopathology, other than one case of mild nuclear pleomorphism observed in Reference Area 1, no other hepatic lesions associated with chemical toxicity were detected. This included observations for megalocytic hepatitis, foci of cellular

alteration, carcinoma, cholangioma, cholangiofibrosis or hydropic vacuolation. The frequencies of macrophage aggregates in livers of fish from the various Areas were low and no cases of moderate to high aggregation were observed. One fish from the Southern portion of the Study Area and one fish from each of Reference Areas 1 and 3 also exhibited a mild inflammatory response. Liver tissues of some fish contained myxosporean parasites but no differences between the Northern and the Southern portion of the Study Area and the pooled Reference Areas were found.

With respect to studies on gill microstructures, X-cells, which are parasitic protozoans, were observed in three plaice from the Reference Areas. Slight thickening of the epithelium of secondary lamellae, or mild hyperplasia, appears to be common within or between Reference and Study Areas. However, microstructural changes which have been associated with chemical toxicity such as severe hyperplasia, epithelial lifting, extensive gill oedema, telangiectasis and lamellar fusion were absent or found at very low frequencies in all Areas.

## 7.0 Discussion

### 7.1 Sediment Component

Evidence of project effects, particularly from drilling and discharge of drill cuttings, in the White Rose EEM program can come from:

- changes in relationships between sediment variables and distances from the drill centres after drilling began; and
- correlations between biological variables (responses) and drilling mud tracers (barium and  $>C_{10}-C_{21}$  HCs).

#### 7.1.1 Physical and Chemical Characteristics

Sediments at White Rose were uniformly sandy, with low fines and gravel content. Fines content in 2000, 2004 and 2005 was usually 1 to 2% and has rarely exceeded 3%. These fines levels are similar to fines levels at Terra Nova (Petro-Canada 2005). Gravel content in White Rose sediments was lower than gravel content at Terra Nova.

The TOC content in White Rose sediments was also low, usually less than 1 g/kg or 0.1%. TOC values of 1% are considered typical of uncontaminated marine sediments (CCME 2006), although this value may be more typical of nearshore rather than offshore sediments. In 2005 and 2000, but not 2004, TOC and fines content in White Rose sediments were significantly positively correlated. Organic carbon is normally associated with finer particles in sediments, but this relationship was weak for White Rose sediments because of the restricted range of fines and TOC content.

There was clear evidence of project effects from drilling and discharge of drill cuttings on concentrations of  $>C_{10}-C_{21}$  HCs and, to a lesser extent, on barium concentrations. Both substances are major constituents of drilling muds, and elevated concentrations would be expected where these muds are used and cuttings discharged.

In 2000, prior to drilling,  $>C_{10}-C_{21}$  HC concentrations at all 46 stations sampled were less than EQL (0.3 mg/kg). In 2004 and 2005,  $>C_{10}-C_{21}$  HC concentrations at stations located 10 or more km from active drill centres were also near or below EQL, but most concentrations within 10 km of active drill centres were greater than EQL. Therefore, concentrations above EQL can be considered evidence of contamination from drilling and, specifically, the use of SBMs. In 2004,  $>C_{10}-C_{21}$  HC concentrations decreased significantly with increasing distances from the Northern and Southern drill centres, after drilling began at these two centres.  $>C_{10}-C_{21}$  HC concentrations did not decrease with increasing distance from the Central drill centre in 2004, but did in 2005, after drilling began at this centre. Distance gradients were steep in both years, with concentrations decreasing by 100 to 1,000 fold over 10 km. The estimated zone of influence for  $>C_{10}-C_{21}$  HCs in 2005 was between 6 and 7 km from the nearest drill centre. Overall concentrations also progressively increased over time.

Results of field monitoring were generally consistent with predictions from a pre-drilling dispersion model (Hodgins and Hodgins 2000). The model predicted that drill cuttings and HCs could be dispersed up to 9 km from drill centres, but that distance gradients would be steep, with most cuttings and HCs deposited near drill centres.

Barium, as barium sulphate (barite), is a major constituent of WBMs and SBMs. Barium occurs naturally in White Rose sediments at concentrations ranging from approximately 120 to 210 mg/kg. Therefore, low-level contamination from drilling can be difficult to detect. Despite this limitation, barium concentrations decreased significantly with distances from the Southern and Central drill centres after drilling began at these two centres. There was no evidence of effects from the Northern drill centre after drilling began at this centre. Overall barium concentrations from stations sampled in all sample years have progressively increased over time. The estimated zone of influence for barium in 2005 was between 2 and 3 km from the nearest drill centre.

In 2005,  $>C_{10}-C_{21}$  HC and barium concentrations, were greater to the Southeast within 1 km of the Central and Southern drill centres, in the direction of the residual current. This small directional effect is consistent with current records at White Rose for 2003 and 2004 (Husky Energy 2005) and with Hodgins and Hodgins (2000), who note that currents at White Rose are generally dominated by wind and tide, with a weak mean flow to the South.

Overall,  $>C_{10}-C_{21}$  HCs were a better indicator of drilling activity for White Rose than barium. However, this conclusion is specific to the White Rose, Terra Nova and other recent offshore oil developments where SBMs based on  $>C_{10}-C_{21}$  HCs are used. In these cases,  $>C_{10}-C_{21}$  HCs provide a specific “fingerprint” of contamination from drilling that can be easily distinguished from background.

Elevated concentrations of HCs and barium have been observed near drill centres and platforms in other offshore oil developments (Table 7-1). Levels of HCs and barium at White Rose within 5 km of the drill centres were within the range noted elsewhere.

**Table 7-1 Hydrocarbon and Barium Concentration at White Rose and at Other Development Sites**

Well Location	Year of Study	Distance from Source (m)	Total Petroleum Hydrocarbon (mg/kg)	Barium (mg/kg)
White Rose (Husky Energy 2001, 2005)	2005	300 to 750	<3 to 261.7	210 to 810
		750 to 2,500	<3 to 54.6	140 to 380
		2,500 to 5,000	<3	150 to 220
	2004	300 to 750	8.74 to 275.92	190 to 1,400
		750 to 2,500	0.21 to 21.95	120 to 470
		2,500 to 5,000	<3 to 6.60	140 to 230
	2000	300 to 750	<3	140 to 180
		750 to 2,500	<3	140 to 190
		2,500 to 5,000	<3	140 to 210

Well Location	Year of Study	Distance from Source (m)	Total Petroleum Hydrocarbon (mg/kg)	Barium (mg/kg)
Terra Nova (Petro-Canada 1998, 2001, 2002, 2003, 2005)	2004	140 to 750 750 to 2,500 2,500 to 5,00	7.78 to 6,580 2.9 to 72.2 <3 to 4.3	140 to 2,100 100 to 340 63 to 190
	2002	140 to 750 750 to 2,500 2,500 to 5,000	<3 to 931 <3 to 49 <3 to 4.8	110 to 2,200 84 to 330 83 to 200
	2001	750 to 2,500 2,500 to 5,000	<3 to 29.5 <3 to 8.13	100 to 190 87 to 180
	2000	750 to 2,500 2,500-5,000	0.59 to 14.4 <3 to 5.59	92 to 210 80 to 230
	1997	750 to 2,500 2,500-5,000	<32.5 <32.5	87 to 190 79 to 280
Gulf of Mexico (NPO-895) (Candler et al. 1995)	1993	50 200 2,000	134,428 80 to 11,460 24	47,437 542 to 5,641
Gulf of Mexico (MAI-686) (Kennicutt et al. 1996)	1993	200	40	1,625
		500	43	1,134
		3,000	49	1,072
Gulf of Mexico (MU-A85) (Kennicutt et al. 1996)	1993	200	42.3	3,706
		500	31.7	1,817
		3,000	27.1	1,094
Gulf of Mexico (HI-A389) (Kennicutt et al. 1996)	1993	200	65	13,756
		500	33	3,993
		3,000	32	1,293
North Sea (Beatrice) (Addy et al. 1984)	1982	250	8 to 759	
		750	5 to 105	
		3,000	3 to 73	
Dutch Continental Shelf (K14-13) (Daan and Mulder 1996)		200	54 to 161	
Norway (Valhall) (Hartley 1996)	1985	250		19,000 to 96,000
		500		3,700 to 9,300
		3,000		280 to 430
North Sea (Brent) (Massie et al. 1985)	1981	800	41 to 61	
		3,200	33 to 43	
North Sea (Forties) (Massie et al. 1985)	1980	800	9 to 78	
		3,200	16 to 55	
Gulf of Mexico (Matagorda 622) (Chapman et al. 1991; Brooks et al. 1990)	1987	25	757 ±1,818	6,233
		150		12,333
		750		980
		3,000		
Santa Maria Basin (Hidalgo) (Phillips et al. 1998)	1991	125		1,250
		500		975
		1,000		1,050
Norway (Ekofisk) (Ellis and Schneider 1997)	1996	750		3,650
		2,000		2,214
		5,000		667
Norway (Gyda 2/1-9) (Bakke et al. 1995)	1994	100 to 200	236	
Norway (Tordis) (Gjøs et al. 1991)	1990	500	8,920	
Norway (U/a 2/7-29) (Vik et al. 1996)		200	1,000 to 2,368	
North Sea (UK) (UKOOA 2001)	1975 to 1995	0 to 500	124 to 11,983	84 to 2,040
		>500 to 2,000	3 to 164	7 to 1595
		>2,000 to 5,000	3 to 76	8 to 729

Note: - Absolute barium levels should not be compared across projects because of potential difference in measurement techniques (Hartley 1996), and differences in background levels.

In 2005, redox levels increased with distances from the Southern and Central drill centres, but decreased with distance from the Northern drill centre. The zone of influence for redox levels extended to between 2 and 3 km from source. Reduced redox levels were also observed near drill centres at Terra Nova (Petro-Canada 2005), after several years of drilling. Reductions in redox levels may be a delayed effect, resulting from breakdown of HCs.

Sulphur, as barium sulphate, is a constituent of WBMs and SBMs, but there are also many natural sources of sulphur. In 2004, but not in 2005, there was some evidence of decreases in sulphur concentrations with distance from the drill centres. In both years, sulphur concentrations were significantly positively correlated with barium and  $>C_{10}-C_{21}$  HC concentrations. Sulphur concentrations at most stations have varied within a narrow range (0.02 to 0.04%, or 200 to 400 mg/kg). Consequently, any distance gradients would be weak, and detectable contamination would be restricted to stations near (less than 1 km from) drill centres.

Fines content increased with increasing depth in all three sample years (2000, 2004 and 2005). Finer particles are expected to move down-slope. There has been no evidence of a general increase in fines content near drill centres.

TOC content decreased with distance from the Central drill centre in all three sample years, and was unrelated to depth or distances from the other two drill centres.  $>C_{10}-C_{21}$  HC contamination would only affect TOC levels at high concentrations. Most  $>C_{10}-C_{21}$  HC concentrations in 2004 and 2005 were less than 10 mg/kg.

Concentrations of metals other than barium were unaffected by drilling. Ammonia concentrations were also unaffected by drilling.

In summary, there was clear evidence that drilling elevated concentrations of barium and  $>C_{10}-C_{21}$  HCs, two major constituents of drilling muds. In 2005, barium contamination extended 2 to 3 km from source (i.e., drill centres), and  $>C_{10}-C_{21}$  HC contamination extended 6 to 7 km from source. Redox levels were also reduced near the Central and Southern drill centres. Reduction in redox levels extended to 2 to 3 km from source. Sulphur levels may also be elevated near drill centres, but overall distance gradients would be weak and difficult to detect. Levels of fines, TOC, metals other than barium, and ammonia did not appear to be affected by drilling.

## 7.1.2 Biological Effects

### 7.1.2.1 Project Effects

None of the 146 sediment samples collected in 2000, 2004 and 2005 were toxic to bacteria in laboratory tests. One of the 44 samples tested in 2005 was toxic to amphipods in laboratory tests, and survival was reduced to less than 70% in another 2005 sample. Otherwise, survival has always been greater than 70% and usually greater than 80%. The two 2005 samples with reduced survival represented only two of the many stations where effects on *in situ* invertebrate communities were observed.

In 2005, total abundance, polychaete dominance and amphipod abundance decreased with increasing  $>C_{10}-C_{21}$  HC concentrations. Total abundance and polychaete dominance were reduced near the Southern drill centre and amphipod abundance was

reduced near all drill centres. Total abundance and polychaete dominance were closely related because polychaetes accounted for approximately 75% of the invertebrates collected. Therefore, reductions in abundances of polychaetes would reduce both total abundance and polychaete dominance.

The reductions in polychaete dominance suggest that bivalves, the other dominant organisms (17% of total abundance), were unaffected by drilling or less affected than polychaetes. Reductions in polychaete abundances were unexpected, since polychaetes are typically considered tolerant organisms (e.g., Peterson et al. 1996). Green and Montagna (1996) showed that polychaete:amphipod ratios increased near drilling platforms in the Gulf of Mexico. There was also some evidence from the Terra Nova EEM program that drilling and HCs may have stimulatory effects on some polychaetes at that location (Petro-Canada 2005).

There appeared to be a lag between the onset of drilling and effects on polychaetes. Increases in total abundance with distance from the Southern drill centre were much stronger in 2005 than in 2004. Similarly, polychaete dominance was unrelated to distance from the Southern drill centre in 2004, but increased significantly with distance in 2005. Neither variable appeared to be affected by drilling at the Central drill centre, which began in 2005. The two variables also appeared unaffected by drilling at the Northern drill centre, which began in 2004. However, it should be noted that relationships between the two variables and  $>C_{10}-C_{21}$  HC concentrations in 2005 suggested that concentrations greater than EQL near the Central and Northern drill centres had some effects, even if these effects were not evident from overall distance relationships.

Relationships between total abundance and polychaete dominance versus  $>C_{10}-C_{21}$  HC concentrations also increased in strength between 2004 and 2005. In 2004, estimated threshold  $>C_{10}-C_{21}$  HC concentrations for effects were 2.2 mg/kg for total abundance and 37 mg/kg for polychaete dominance. In 2005, a threshold concentration could not be estimated for total abundance because decreases with increasing concentration occurred across the entire concentration range (i.e., any threshold would be less than the EQL of 0.3 mg/kg). The estimated threshold concentration for effects on polychaete dominance in 2005 was 0.9 mg/kg, or 2% of the 2004 threshold. In other words, effects on polychaetes were observed at lower  $>C_{10}-C_{21}$  HC concentrations in 2005 than in 2004.

Most polychaetes are small and relatively short-lived, and would be expected to respond rapidly to drilling and other anthropogenic discharges and stressors. Effects may not have been evident immediately after drilling began because total abundance and polychaete dominance reflect combined effects on many taxa. These effects could differ in magnitude or even direction, and also timing, among taxa, and indirect effects from alterations of abundance of competitors or predators could also occur. For example, in the Terra Nova EEM program, the dominant Spionidae appeared to be replaced by Paronidae and several other polychaete families at the most contaminated near-field station(s) (Petro-Canada 2005). With both Spionidae and Paronidae abundant at White Rose, gradual replacement of one taxon by the other would dilute and delay effects on total abundance and polychaete dominance. In future years, effects on specific polychaete families will be assessed.

Amphipods responded more rapidly to drilling, possibly because they are a relatively homogeneous group of small, short-lived organisms (see below). Effects from the Southern and Northern drill centres were evident in 2004 after drilling began at these two centres, and effects from the Central drill centre were evident in 2005, after drilling began at this centre. The effects of distances from the Northern and Southern drill centres did not change significantly between 2004 and 2005, which suggests that effects did not intensify over time. However, relationships between amphipod abundance and  $>C_{10}-C_{21}$  HC concentrations were stronger (i.e., threshold concentrations were lower) in 2005 than in 2004, which suggests some intensification of effects.

Amphipods are sensitive to drill cuttings discharges, as this study and others indicate (e.g., Peterson et al. 1996). At White Rose, there were also several largely serendipitous factors that made effects easier to detect than for other taxa. Amphipod abundance was a function of abundances of a few taxa (families) within a single Order that may respond similarly to drilling and other activities. Amphipods were also abundant enough to analyze using parametric methods, with abundances of 0 occurring only in 2005 and only near drill centres. Natural distance gradients (*decreases* in amphipod abundance with distance, observed during baseline sampling) were the opposite of observed gradients (*increases* with distance during EEM years), and depth effects were negligible, which removed two possible confounding factors.

Estimated zones of effects for total abundance and polychaete dominance in 2005 were between 2 and 3 km from source. These zone of effects are underestimates, since effects on both variables were observed across all or most of the range of detectable  $>C_{10}-C_{21}$  HC concentrations. Effects on amphipods extended to all but the most distant stations (i.e., to 5 or more km) and a zone of effects limit was not quantified because this estimate would not be robust. Effects on amphipods extended across the entire range of detectable  $>C_{10}-C_{21}$  HC concentrations.

Decreases in numbers or relative abundance within these zone of effects were approximately 65% for total abundance, 20% for polychaetes and 55% for amphipods. Table 7-2 shows minima and maxima for the three affected benthic invertebrate variables. Using these values, decreases in total abundance since baseline within 2.5 km of drill centres ranged from 50 to 80%. Decreases in the relative abundance of polychaetes within 2.5 km of drill centres ranged from 0 to 35%. While for amphipods, decreases within 5 km of drill centres range from 45 to 60%.

**Table 7-2 Minimum and Maximum Values by Distance Class for Total Abundance, the Relative Abundance of Polychaetes and Total Abundance of Amphipods**

Year	Distance Class (m)	Total Abundance (#)	Relative Abundance of Polychaetes (%)	Total Abundance of Amphipods (#)
2000	300 to 750	500 to 918	73 to 82	15 to 44
	750 to 2,500	401 to 1152	54 to 86	12 to 53
	2,500 to 5,000	507 to 1198	66 to 81	5 to 46
	> 5000	322 to 1040	69 to 89	7 to 36
2004	300 to 750	262 to 429	50 to 80	0 to 14
	750 to 2,500	312 to 680	66 to 79	2 to 30
	2,500 to 5,000	304 to 810	58 to 86	6 to 35
	> 5000	291 to 847	50 to 87	6 to 33



Year	Distance Class (m)	Total Abundance (#)	Relative Abundance of Polychaetes (%)	Total Abundance of Amphipods (#)
2005	300 to 750	75 to 316	30 to 61	0 to 22
	750 to 2,500	87 to 581	34 to 87	0 to 22
	2,500 to 5,000	181 to 772	59 to 86	2 to 25
	> 5000	226 to 1197	54 to 85	8 to 55

Note: - This table provides an indication of the magnitude of effects and variance at various distances. This complements zone of effects models in Section 5

The effects of the White Rose development on invertebrates, especially amphipods, were spatially more extensive and occurred at lower HC concentrations than in most other studies of offshore oil development effects. For example, effects from North Sea developments have generally extended 1 to 3 km from oil platforms (Olsgård and Gray 1995; Daan et al. 1994, 1996); effects in the Gulf of Mexico were generally not evident more 1 km from platforms (Green and Montagna 1996). Similarly, effects tend to be observed at HC concentrations greater than 10 to 100 mg/kg, although Kingston (1992) notes that some sensitive taxa may be affected at concentrations less 10 mg/kg.

Some caution is required in inferring that effects of the White Rose development on invertebrates were worse than at other development sites. There are important differences in drilling programs (e.g., platforms versus drill centres; drilling muds used), study areas and sampling designs, field and laboratory methods, and invertebrate communities and community variables analyzed among studies that complicate comparisons among studies.

The White Rose study design and data analyses were powerful enough to detect some relatively small effects. The sampling design and associated data analyses are unique to the White Rose and Terra Nova projects, and were deliberately selected to better define the spatial extent of effects (i.e., zone of influence/effects). Several other factors unique to the White Rose area also made effects easier to detect (e.g., see above discussion of effects on amphipods). Sediments were relatively homogeneous, which limited any confounding effects of variance in fines or TOC content. Depth effects, although present, were relatively small.  $>C_{10}-C_{21}$  HCs were an excellent tracer of contamination from SBMs and predictor of biological effects, because concentrations were unaffected by background variance and EQL were lower than for several other gross HC measures (e.g., see EQL for Total Petroleum Hydrocarbons (TPH) in Table 7-1).

Kingston (1992) noted that decreases in diversity were generally observed where HC concentrations near oil platforms reached 50 to 60 mg/kg, and there were HC levels greater than this in the 2004 and 2005 White Rose programs. However, analyses of relationships between diversity and either distance or  $>C_{10}-C_{21}$  HC concentrations in this report provided no evidence of project effects on diversity. Although other analyses were powerful enough to detect some subtle depth and gravel effects on diversity, despite naturally low diversity at White Rose (see Section 7.1.2.2).

Peterson et al. (1996) noted that richness may be reduced near drill cuttings and other discharges, with the reductions reflecting adverse effects on a broad range of rarer taxa. There was no evidence of effects on richness at White Rose, although richness (typically 20 to 30 families per station) was relatively high, given the low diversity and dominance of a few families. Most rare taxa may be unaffected by drilling.

Other studies and authors have also noted that taxa other than amphipods (e.g., some echinoderms and bivalves) may be sensitive to and affected by drilling (Kingston 1992; Daan et al. 1994; Peterson et al. 1996). At 146 stations sampled at White Rose over three years, the maximum number of echinoderms collected per station was 21. Echinoderm abundances are the sum of abundances of a broad range of taxa (starfish, sea urchins, sea cucumbers and relatives) within the Phylum Echinodermata. These taxa may not respond similarly to drilling, but abundances are too low to allow analyses at lower taxonomic levels (e.g., Class or Order). The dominant bivalve family was Tellinidae (mostly *Macoma calcarea*). *Macoma* species are commonly used in laboratory studies to assess bioaccumulation of metals and organic compounds, partly because they are tolerant enough to survive and continue respiring and feeding at relatively high contaminant levels.

Elevated barium concentrations are unlikely to be the direct cause of observed effects on polychaetes and amphipods. Effects (i.e., relationships with distance or  $>C_{10}-C_{21}$  HCs) occurred within the background range of barium concentrations (120 to 210 mg/kg). Barium, as barite in fine particulates, is primarily a physical irritant rather than a chemical toxicant, adversely affecting cilia and gills (Barlow and Kingston 2001; Armsworthy et al. 2005). Gray et al. (1990) suggested that effects from HC in oil-based drilling muds were greater than any effects of barium in the North Sea. These authors noted that metal impurities in WBMs may have effects where barium concentrations are high, but concentrations of metals in White Rose sediments were below sediment quality guidelines (Section 7.1.3) and unrelated to invertebrate community variables.

Laboratory toxicity tests with amphipods indicate that effects do not occur at  $>C_{10}-C_{21}$  HC concentrations less than 1,900 mg/kg (Payne et al. 2001), well above any concentrations measured in White Rose sediments; and, as noted above, only one White Rose sample was found to be toxic to amphipods in laboratory tests in 2005. *In situ*, estimated thresholds for effects on polychaetes and amphipods in 2005 were near or below EQL (0.3 mg/kg), or approximately three orders of magnitude below the laboratory effects threshold. Given the differences between field measurements and laboratory measurements, reduced field abundances are not likely due to direct acute toxicity. Rather, community effects could be due to indirect effects or to chronic toxicity involving longer term exposure.

Other physical and chemical characteristics measured in the White Rose EEM program either do not occur at concentrations associated with biological effects (Section 7.1.3), or did not vary with either distances from the drill centres or biological responses.

In summary, there was clear evidence of adverse drilling effects on polychaetes and amphipods from field surveys of invertebrate communities. At this stage in the life of the White Rose field, these effects appeared to be spatially more extensive and occur at lower HC concentrations than in other studies. Reductions in polychaete abundance and dominance were unexpected, since polychaetes are typically considered tolerant organisms and drilling can have stimulatory effects on these organisms. Overall, biological effects of drilling at White Rose did not follow the model developed by Peterson et al. (1996) (simplistically, drilling increased polychaete:amphipod ratios, and sometimes reduced diversity/richness and echinoderm abundance) based on an extensive review of field monitoring programs.

Barium and other physical and chemical characteristics measured in the White Rose EEM program are unlikely to be the cause of observed biological effects on invertebrate communities. These effects were strongly correlated with  $>C_{10}-C_{21}$  HC concentrations, with estimated effects concentrations much lower than reported in other field studies or in laboratory toxicity tests of the effects of SBMs.

In the short term, future monitoring years are required to verify that the unique results observed to date in the White Rose EEM program are real and persistent over time. Questions of interest are:

- which polychaete families are affected, either positively or negatively?
- are taxa other than polychaetes and amphipods affected?
- will effects continue to increase or intensify as they did between 2004 and 2005, or will then plateau or decrease?

In the long term, assessing potential enrichment effects from microbial degradation of HCs, recovery from current effects, and potential effects of produced water discharges will be important issues.

#### 7.1.2.2 Effects Unrelated to the Project

Fines and TOC content in White Rose sediments had little or no effect on benthic invertebrate community variables, probably because fines and TOC content were low and did not vary widely. Over all three sample years (2000, 2004 and 2005), richness and diversity increased, and polychaete dominance decreased, with increasing gravel content and depth. These gravel and depth effects were relatively small, but are of interest because:

1. The ability of the program to detect these small effects implies that project effects on richness and diversity would have been detected if they occurred (project effects on polychaete dominance were detected).
2. The emphasis on particle size effects in other studies has typically been on fines (e.g., “smothering” from discharge of fine drill cuttings), but gravel appears to be the more important particle size variable in the predominantly sandy sediments on the Grand Banks.
3. Depth effects occurred over a narrow depth range, and there is no apparent physical factor (e.g., variation in light or temperature) that would explain these effects.

Higher gravel content can positively affect richness and diversity by increasing habitat or “niche” diversity (i.e., the number of interstitial spaces, and the variance in the size of these spaces). At Terra Nova, where gravel content was greater and more variable than at White Rose, gravel effects were much stronger (Petro-Canada 2005). For both White Rose and Terra Nova, variance in fines content less than 5% and usually between 1 to 2% was unlikely to have any physical effects (e.g., smothering) on invertebrates.

Depth effects on richness, diversity and polychaete dominance were largely independent of gravel and fines content, although these two particle size variables generally increased with depth. The depth effects occurred over a narrow range of depths from 115 to 140 m (the effects of the two reference stations with extreme depths of 108 and 175 m were removed or reduced by excluding these stations or analyzing depth ranks). Polychaetes were more dominant at Terra Nova (shallower depth range: 90 to 100 m) than at White Rose (Petro-Canada 2005). However, richness and diversity were not greater at White Rose than at Terra Nova. Therefore, depth gradients for richness and diversity appear to be localized and specific to White Rose. Total abundance also increased with increasing depth, although polychaete dominance decreased. Therefore, positive depth effects on abundances of non-polychaetes (e.g., the bivalves Tellinidae) were probably much stronger than indicated by analyses of summary measures that included polychaetes.

Assuming that variation in pressure, light and temperature with depth was minimal, and that depth effects were largely independent of particle size, some other unmeasured factor was responsible for depth effects. The depth effects observed at White Rose are unusual, because total abundance and/or richness usually decrease with increasing depth (e.g., Paine et al. 1996; Wilson et al. 1999). Depth effects were relatively small, and can be reduced or removed by including depth as an additional X variable in regression and correlation analyses.

### 7.1.3 CCME Guidelines

The Canadian Council of Ministers of the Environment (CCME) provides marine sediment quality guidelines for PAHs and several metals (CCME 2006). Interim Sediment Quality Guidelines (ISQG) are Threshold Effects Levels (TEL) below which biological effects are rarely observed. Probable Effects Levels (PEL) are levels above which effects are often observed. The CCME guidelines are based on literature reviews of concentration-effects relationships from laboratory and field studies (i.e., co-occurrence or correlation of chemical contamination and biological effects).

Table 7-3 compares maximum levels of PAHs and metals in White Rose sediments to CCME ISQG and PEL. No PAHs were detected at EQLs of 0.05 mg/kg, and these EQLs were less than PEL. However, EQLs were higher than ISQG for acenaphthene, acenaphthylene, anthracene, dibenz(*a,h*)anthracene, fluorene, 2-methylnaphthalene and naphthalene. Maximum concentrations and EQLs for the seven metals with guidelines were well below ISQG. At these low levels, most metals would be essential elements rather than toxicants.

**Table 7-3 Comparison of Measured Concentrations of PAHs and Metals to Canadian Sediment Quality Guidelines**

Variable	ISQG (mg/kg)	PEL (mg/kg)	Maximum value		
			2000 (n=46 stations)	2004 (n=56 stations)	2005 (n=44 stations)
Acenaphthene	0.00671	0.0889	<0.05	<0.05	<0.05
Acenaphthylene	0.00587	0.128	<0.05	<0.05	<0.05
Anthracene	0.0469	0.245	<0.05	<0.05	<0.05
Benz(a)anthracene	0.0748	0.693	<0.05	<0.05	<0.05
Benzo(a)pyrene	0.088	0.763	<0.05	<0.05	<0.05
Chrysene	0.108	0.846	<0.05	<0.05	<0.05
Dibenz(a,h)anthracene	0.00622	0.135	<0.05	<0.05	<0.05
Fluoranthene	0.113	1.494	<0.05	<0.05	<0.05
Fluorene	0.0212	0.144	<0.05	<0.05	<0.05
2-Methylnaphthalene	0.0202	0.201	<0.05	<0.05	<0.05
Naphthalene	0.0346	0.391	<0.05	<0.05	<0.05
Phenanthrene	0.0867	0.544	<0.05	<0.05	<0.05
Pyrene	0.153	1.398	<0.05	<0.05	<0.05
Arsenic	7.24	41.6	2	<2	<2
Cadmium	0.7	4.2	<0.05	0.08	0.07
Chromium	52.3	160	4	7	5.5
Copper	18.7	108	4	3	2.9
Lead	30.2	112	5.1	4.0	5.9
Mercury	0.13	0.7	<0.01	<0.01	<0.01
Zinc	124	271	14	9	10

Notes: - Source – CCME (2006); [www.ccme.ca/ccme](http://www.ccme.ca/ccme)  
 - CCME guidelines are not available for other variables measured at White Rose

## 7.2 Commercial Fish Component

### 7.2.1 Biological Characteristics

Analysis of crab Biological Characteristics (size and frequencies of recent moult) in 2004 and 2005 indicated that Study Area and Reference Area crab were similar. Most of the variance in Biological Characteristics occurred at small spatial scales, among trawls or composites within Areas. In 2004, crab from Reference Area 4 were larger, and frequencies of recent moult much lower, than in other Areas. In 2005, only one composite of eight crab was collected in Reference Area 4, and those few crab did not appear unusual relative to crab from other Areas.

Carapace width (i.e., size) of crab used in body burden analyses decreased by 30% between 2004 and 2005. This size reduction was greater than most size differences among Areas and among composites within Areas, but had little effect on differences in body burdens between the two years (Section 7.1.2).

In 2005, plaice from Reference Area 3 used in body burden and health analyses were all large mature females. Consequently, composite mean weights were approximately double those in other Areas, where composites consisted of a mix of large mature females and smaller males and immature females. The large size difference between Reference Area 3 and other Areas had some effects on body burdens, but effects of smaller size differences among and within the other Areas were also important. Therefore, size should continue to be used as a covariate in analyses of body burden.

Study Area composite mean weights were within the Reference Area range in both 2004 and 2005, partly because the Reference Area range was wide in 2005 and, to a lesser extent, in 2004.

### **7.2.2 Body Burden**

Metal concentrations in crab claws, plaice livers and plaice fillets from the Study Area were similar to or lower than Reference Area concentrations in both 2004 and 2005. HCs have not been detected in crab claw. HCs have only been detected in one plaice fillet, from Reference Area 4, in 2005; and the chromatogram for this sample did not indicate the presence of drill fluids. HCs were detected in every liver sample, except one 2004 sample with unusually high EQL. These HCs did not resemble drill fluid and peaks observed on chromatograms were consistent with those expected for extracted fatty acids.

There were few consistent and significant differences in metal and HC concentrations in crab and plaice tissue across both EEM years (2004 and 2005). Copper and zinc in plaice liver were minor exceptions, with concentrations slightly but significantly lower in Study Area fish. Study versus Reference Area differences in body burdens also have not changed between years, largely because they were small in both years.

### **7.2.3 Taste Tests**

There was no taste difference between the Reference and Study Areas for crab. For plaice, panelists showed a preference for Reference Area samples in the hedonic scaling test but could not distinguish between Study and Reference Area samples in the triangle tests. For both plaice and crab, there were no consistent comments from panelists identifying abnormal or foreign odour or taste. For plaice, most panelists preferred the texture of Reference Area samples. Combined, test results do not indicate the presence of taint in either crab or plaice at White Rose in 2005.

### **7.2.4 Fish Health Indicators**

#### **7.2.4.1 Haematology**

There were no apparent qualitative differences in morphology or staining characteristics of red blood cells in samples from the different Areas.

Differential blood cell counts were similar in fish from all of the Reference Areas as well as in fish from the southern portion of the Study Area. However, blood samples of fish from the northern portion of the Study Area had fewer lymphocytes and more thrombocytes. Although statistical differences were obtained for these two cell types, it is important to note that the changes in cell numbers were quite small. Such small changes can be attributed to natural variation (De Pedro et al. and references therein, 2005).

#### **7.2.4.2 Mixed Function Oxygenase**

Since maturity stage might result in some loss of sensitivity for resolving contaminant mediated differences in females during spawning (e.g., Mathieu et al. 1991; Whyte et al. 2000), MFO enzyme activities were analyzed separately in immature and mature female American plaice from the different Areas. There were no significant differences in MFO

enzyme activities in either mature females or immature females among Reference Areas, between the southern and northern portion of the Study Area or between the Study Area and combined Reference Areas. Similar results were obtained with males (all maturity stages pooled).

#### 7.2.4.3 Pathology

Gross pathology was assessed visually in all fish during the necropsies for any external or internal abnormalities or parasites. One fish from Reference Area 1 exhibited a skin lesion while one fish from each of Reference 1 and 4 displayed gill achromasia (pale gill filaments) which were confirmed by microscopy examination to be X-cell lesions.

With respect to liver histopathology, other than one case of nuclear pleomorphism observed in Reference Area 1, no other hepatic lesions associated with chemical toxicity in field and laboratory studies (e.g. Myers and Fournie 2002) were detected. This included observations for megalocytic hepatosis, eosinophilic, basophilic and clear cell foci, high aggregation of macrophages, carcinoma, cholangioma, cholangiofibrosis and hydropic vacuolation.

However, a few hepatic differences were noted. A mild inflammatory response was observed in one fish from Reference Area 1, one fish from Reference Area 3 and one fish from the southern portion of the Study Area. As noted in previous years, a “patchy distribution” of hepatocellular vacuolation, not associated with degenerative changes, was observed in a few fish from both Areas and is likely linked to gonadal maturation (Timashova 1981; Bodammer and Murchelano 1990; Couillard et al. 1997). Also, liver tissues of some fish contained myxosporean parasites but no differences between Areas were found. The infestation did not appear to result in any other pathological changes in hepatic tissues.

Observations on mild inflammatory responses, hepatocellular vacuolation and parasitism are of value in relation to providing general information on their presence in the area. However, it is important to note, from an EEM perspective, that liver lesions associated with chemical toxicity were absent or found only at a very low prevalence in the general area.

With respect to studies on gill microstructures, slightly significant differences in the general thickening of the secondary lamellae epithelium (measured as distal, tip or basal hyperplasia) were observed between Areas. These differences, noted this year as well as in previous years, indicate that a certain degree of variability in the gill lamellar thickening of plaice is of a background nature. However, microstructural changes which could be more pathological such as severe hyperplasia, epithelial lifting, extensive gill oedema, telangiectasis and lamellar fusion (e.g., Mallat 1985) were absent or found at very low frequencies in all areas.

The presence of gill achromasia and X-cell lesions in one plaice from Reference Area 1 and two plaice from Reference Area 4 is of interest. This type of lesion has been reported in various bottom-dwelling fish species, particularly flatfishes and cod living in temperate to cold sea-water (Dethlefsen et al. 1996; Møllergaard and Lang 1999; McVicar et al. 1987). Dessler and Khan (1982) also observed X-cells in the gills of eelpouts from several areas off coastal Newfoundland and Labrador. There had been some debate on whether X-cells are host cells such as protozoa (Alpers et al. 1977) or

cells which have undergone transformation due to pollution or viral infection (Lange and Johannessen 1977; Peters et al. 1978). However it has been confirmed recently that X-cells in fish are parasitic protozoans (Miwa et al. 2004).

As for the liver histopathological indices and from an EEM perspective, it is of interest to note the absence or very low incidence of gill lesions associated with chemical toxicity in all Areas.

Overall, the results obtained on external and internal abnormalities, haematology, hepatic MFO enzymes and liver and gill histopathology indicate that the present health status of plaice collected at the White Rose Study Area is similar to that at the References Areas.

Some variability was noted between a few sites with respect to fish or organ condition but this can be attributed to natural causes such as slight differences in feeding or reproductive status. The slight differences in haematology can equally be attributed to natural variation. Of particular interest was the virtual absence of inter-site variability with respect to the health effect indicators commonly associated with chemical toxicity. This included not only MFO enzymes but also a wide range of liver and gill lesions.

### 7.3 Summary of Effects and Monitoring Hypotheses

As discussed in Section 1, monitoring hypotheses were developed in Husky Energy (2004) as part of EEM program design to test effects predictions and determine physical and chemical zones of influence.

These hypotheses (reiterated in Table 7-4) were set up to guide interpretation of results. As noted in Section 1, the “null” hypotheses ( $H_0$ ) always state that no pattern will be observed.

**Table 7-4 Monitoring Hypotheses**

<b>Sediment Component</b>
$H_0$ : There will be no change in SQT variables with distance or direction from project discharge sources over time.
<b>Commercial Fish Component</b>
$H_0(1)$ : Project discharges will not result in taint of snow crab and American plaice resources sampled within the White Rose Study Area, as measured using taste panels.
$H_0(2)$ : Project discharges will not result in adverse effects to fish health within the White Rose Study Area, as measured using histopathology, haematology and mixed function oxygenase (MFO) induction.

Note: - No hypothesis is developed for plaice and snow crab body burden, as these tests are considered to be supporting tests, providing information to aid in the interpretation of results of other monitoring variables (taste tests and health).

Given results observed in the 2005 EEM program, the null hypothesis is rejected for the Sediment Component of the program, but null hypotheses are not rejected for the Commercial Fish Component. Rejection of the null hypothesis for the Sediment Component was expected since drill cuttings modeling and EIS predictions do indicate that there should be change in SQT variables with distance or direction from discharge sources. The following re-iterates and summarizes project effects.



As indicated above, there was clear evidence that concentrations of  $>C_{10}-C_{21}$  HCs and barium were elevated by drilling activity near drill centres. Redox levels were reduced near the Central and Southern drill centres. There was more equivocal evidence that sulphur concentrations may also have been elevated by drilling. Elevated concentrations of  $>C_{10}-C_{21}$  HCs and barium at White Rose compare to levels observed at other developments.

Sediment contamination did not extend beyond the zone of influence predicted by drill cuttings modeling (Hodgins and Hodgins 2000).  $>C_{10}-C_{21}$  HC contamination extended to between 6 and 7 km from source. Barium contamination extended to 2 to 3 km from source. Reduction in redox levels extended to 2 to 3 km from source. Any contamination from sulphur would be limited to within 1 km from source.

Weak directional effects were noted for both  $>C_{10}-C_{21}$  HCs and barium in 2005, with dispersion primarily to the Southeast within 1 km from the Southern and Central drill centres. This is consistent with current records at White Rose for 2003 and 2004 (Husky Energy 2004), and with Hodgins and Hodgins (2000), who note that currents at White Rose are generally dominated by wind and tide, with a weak mean flow to the south.

One of the 44 samples tested in 2005 was toxic to amphipods in laboratory tests and survival was reduced to 70% in another samples. The two 2005 samples with reduced survival represented only two of the many stations where effects on *in situ* invertebrates were observed.

In field samples, total abundance and polychaete dominance were reduced near the Southern drill centre and decreased with increasing  $>C_{10}-C_{21}$  concentrations. Increases in total abundances with distance from the Southern drill centre were stronger in 2005 than in 2004. Similarly, polychaete dominance was unrelated to distance from the Southern drill centre in 2004 but increased significantly with distance in 2005. The two variables appeared to be unaffected by drilling at the Central and Northern centres in distance regressions. However, relationships between the two variables and  $>C_{10}-C_{21}$  in 2005 suggest that concentrations greater than EQL near the Central and Northern drill centres had some effects, even if these effects were not evident from overall distance relationships. Relationships between total abundance and polychaete dominance versus  $>C_{10}-C_{21}$  concentrations also increased in strength between 2004 and 2005.

Amphipod abundance was reduced near the Southern and Northern drill centres in 2004. Amphipod abundance was reduced near all drill centres (Southern, Northern and Central) in 2005, after drilling started at the Central drill centre. Effects of distance from the Northern and Southern drill centres did not change between 2004 and 2005, which suggests that effects did not intensify over time. However, relationships between amphipod abundance and  $>C_{10}-C_{21}$  concentrations were stronger in 2005 than 2004, suggesting some intensification.

Estimated zones of effects for total abundance and polychaete dominance in 2005 were between 2 and 3 km from source. These zones of effects are underestimates, since effects on both variables were observed across all or most of the range of detectable  $>C_{10}-C_{21}$  HC concentrations. Total abundance decreased to approximately 65% of baseline values within the zone of effects. The relative abundance of polychaetes decreases to approximately 20% of baseline values. The zone of influence for amphipods extended to 5 or more km from source and across the entire range of

detectable  $>C_{10}-C_{21}$  HC concentrations. Amphipods abundance decreased to approximately 55% of baseline values within 5 km of drill centres. Zones of effect for total abundance, polychaete dominance and amphipod abundance exceed EIS predictions.

## **7.4 Summary of Other Relevant Findings**

Total abundance, richness and diversity increased, and polychaete dominance decreased, with increasing depth. These depth effects occurred over a relatively narrow depth range (115 to 140 m for all but two stations).

Richness and diversity increased, and polychaete dominance decreased, with increasing gravel content.

Carry-over effects, or persistent differences among stations unrelated to distance and depth, were not significant for most sediment variables when 2000, 2004 and 2005 were compared, but were significant when only 2004 and 2005 were compared. These results suggest that carry-over effects are important over the short term but not over the long term.

## **7.5 Program Modifications**

### **7.5.1 Considerations for Future EEM Programs**

In addition to those recommendations carried over from the 2004 program (see Section 7.5.2), the following recommendations are provided for future programs.

#### **7.5.1.1 Methods**

Consideration should be given to using glass cups for taste tests, rather than plastic cups.

#### **7.5.1.2 Program Elements**

Effects on benthic invertebrates should be examined in future years to determine if patterns observed to date persist, intensify or moderate. More focused studies should be conducted on abundances of individual dominant polychaete and bivalve families (e.g., Spionidae, Paraonidae, Tellinidae) and possibly echinoderms.

#### **7.5.1.3 Study Design and Data Analysis**

Within years, zones of influence/effects for sediment quality variables should be formally defined using hockey-stick (threshold) relationships, where appropriate. Where threshold relationships do not apply (e.g., amphipods), zone of influence/effects should not be quantified.

Where possible, effect size should be quantified at various distances from source using the hockey-stick approach.

Long-term carry-over effects appear weak for sediment quality variables. Therefore, the same stations do not need to be re-sampled over time and RM analyses do not need to

be used for long-term, multi-year effects assessment. Some alternatives for analyzing all stations sampled were introduced in this report, and could be continued. The best approach would probably be to continue sampling a core set of stations every year, and add other stations (e.g., near existing or new drill centres) to address issues inadequately addressed by RM analyses of the core stations.

$>C_{10}-C_{21}$  HCs should be treated as the primary and most useful drilling mud tracer, and dose-response relationships between  $>C_{10}-C_{21}$  HCs and invertebrate community variables should continue to be examined.

Barium should continue to be measured and a zone of influence for barium defined. However, barium is not a useful dose measure ( $X$  variable). Normalizing barium concentrations to aluminum concentrations has minimal value for effects assessment, and should be dropped as a routine part of the White Rose EEM program.

Multi-year comparisons of correlations between sediment physical and chemical variables other than  $>C_{10}-C_{21}$  HCs, and invertebrate community, should be dropped unless those correlations within one or more sample years become stronger than in the past.

Depth should continue to be included in analyses of distance effects, and it may be useful to adjust for depth effects when estimating the zone of effects for invertebrate community variables.

For the Commercial Fish Component, the Study Area should continue to be split into North and South “sub-areas”, with approximately equal numbers of crab and plaice collected from each sub-area. There were some differences between the two sub-areas, and even if there are no differences, the two sub-areas can always be pooled.

Moisture content, fat content and average size of crab or plaice in composites should be used as covariates in analyses of body burden, when appropriate.

## 7.5.2 Actions Taken on 2004 Recommendations

Recommendation: Effects on benthic invertebrates should be examined closely in future years to determine if the patterns observed in 2004 persist or intensify over time. If these effects persist, more focused analyses of the specific taxa affected should be conducted.

2005 Action: The 2005 results indicated that effects on benthic invertebrates intensified in 2005, and closer examination of effects on specific taxa is recommended for future EEM programs (Section 7.5.1.1).

Recommendation: Changes in fines levels should be examined closely in the future.

2005 Action: Fines were examined. The general increase in fines levels observed between 2000 and 2004 was not observed between 2004 and 2005.

Recommendation: Given high geositional accuracy for sediment sampling, cores should be offset to avoid sampling the same area twice within or across years.

2005 Action: Cores were offset.

Recommendation: In order to better link project discharges to variables measured as part of the EEM program, some samples of treated (if applicable) cuttings from wells drilled with both SBMs and WBMS should be analyzed for particle size and chemistry in the same manner as EEM sediment samples.

2005 Action: Preliminary analyses were carried out in 2005/2006. Additional analyses will be performed more systematically in 2006/2007.

Recommendation: A random subset of EEM stations should be selected for chemistry measurement both at the end of the survey and in conjunction with amphipod toxicity tests. Samples to be analyzed in conjunction with toxicity samples should be held at 4°C in the dark, rather than frozen.

2005 Action: This was done. Ammonia decreased with holding time; ammonia also decreased from initiation to completion of the 10 day amphipod toxicity trials in water overlying test sediments. This analysis will not be redone in future years.

Recommendation: Testing for consistency of results when/if different taxonomists are used for benthic invertebrate identification should continue.

2005 Action: There were no changes in taxonomist between 2004 and 2005.

Recommendation: Because of the poor condition of crab used in taste tests in 2004, crab legs should be either cooked at sea, cooled and frozen or they should be boiled without thawing at the Marine Institute. The logistics of cooking crab at sea will be examined before the 2005 field program.

2005 Action: Crab that had recently molted were excluded from taste analysis. Crab were boiled without thawing at the Marine Institute. These procedures will continue in future programs.

Recommendation: In order to improve the accuracy of comments received from the taste panels, panelists should be instructed that samples are being tested for “uncharacteristic odour or taste” and that grit, cartilage or texture should not be considered in their assessment.

2005 Action: Panelists were so instructed.

Recommendation: Blood smears collected at sea for plaice haematology in 2004 were considered of insufficient uniformity for carrying out reliable differential cell counts (see Section 6.4.4.3). This problem will be overcome in the future by dispensing blood into tubes containing an anticoagulant. This will prevent the blood from clotting and provide more time (up to a couple of hours) to prepare adequate smears and ascertain their quality.

2005 Action: This procedure was performed in 2005 and will continue to be performed.

Recommendation: Since Husky Energy is not currently planning any drilling at the NN and SS drill centres, stations around these drill centres should not be sampled in the 2005 program. Stations to be excluded from the 2005 sampling program are: NN1, NN2, NN3, NN4, NN5, SS1, SS2, SS3, SS4, SS5, and SS6.

2005 Action: These stations were not sampled in 2005 but will be sampled if drilling begins at these drill centres.

Recommendation: If differences in percent recent molt persist between the shallower Reference Area 2 and the deeper Reference Area 4 and remaining Areas, consideration should be given to dropping these two Reference Areas for crab taste tests.

2005 Action: Highest recent molt frequency occurred in Reference Area 3 in 2005; Overall, there was no substantial difference in percent recent molt between the Study Area and the combined Reference Areas.

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