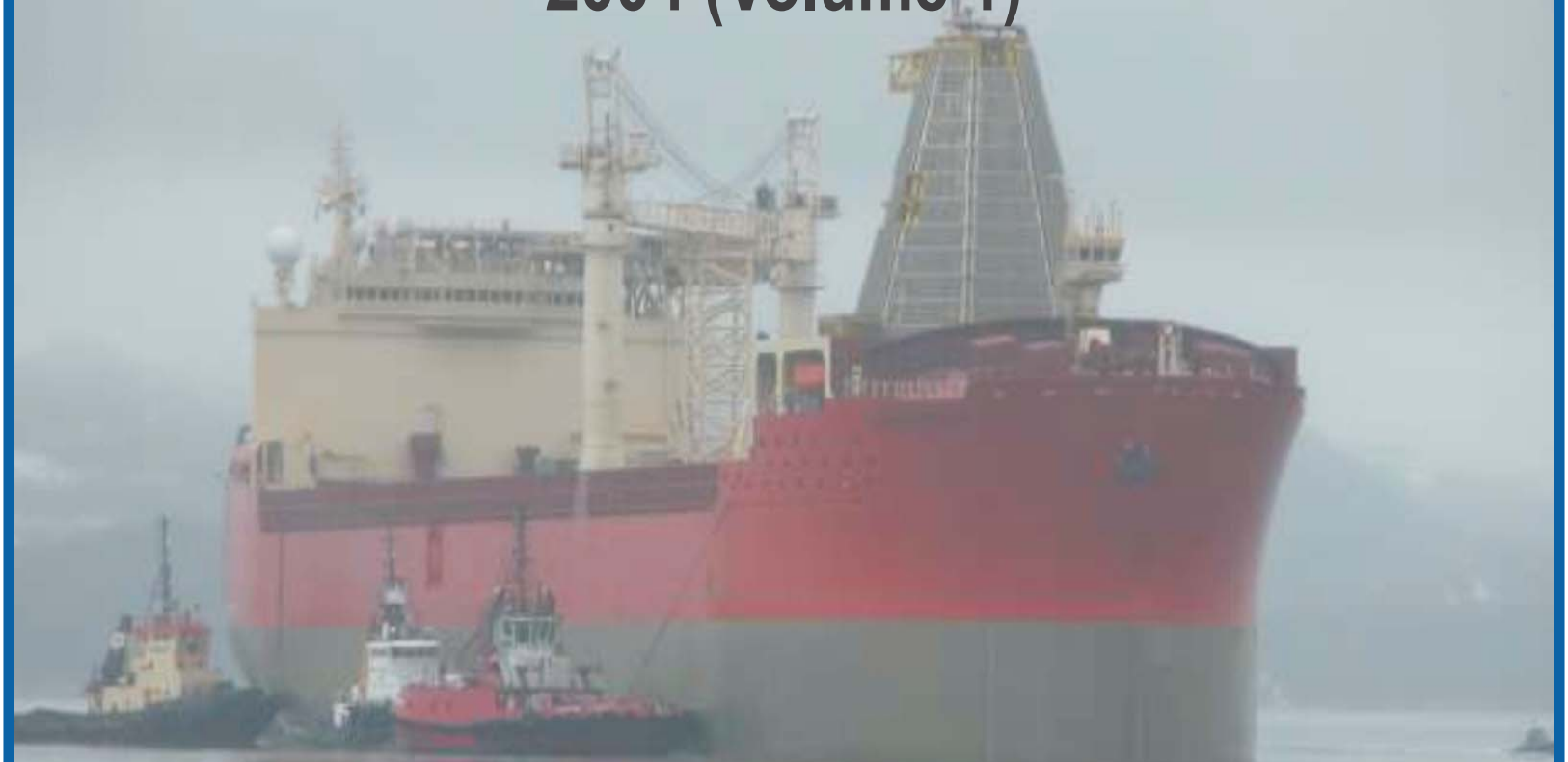
WhiteRose

Husky Energy

White Rose

Environmental Effects Monitoring Program

2004 (Volume 1)



June 2005

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Technical Document

Project:	Location:
White Rose	**East Coast Development**

Document Title:	Total # of Pages:
2004 Environmental Effects Monitoring Program Report	**165**

Document No.:	HDMS IID:	Revision No.:
WR-M-00-C-RP-00011-001	**003936582**	**B1**

Comments:

Revision	Date	Reason For Issue	Prepared	Checked	Approved
B1	16-May-05	Issued for Comments	E.M. DeBlois Jacques Whitford	D.G. Taylor Environmental Coordinator	K. Dyer HSEQ Manager

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 first year of sampling for the EEM program, which was conducted in the summer of 2004. Findings are related to results of sampling conducted under the Baseline Characterization program (Husky Energy 2001; 2003).

In 2004, seafloor sediments were sampled at 31 locations along transect lines radiating from the centre of the development; 14 locations surrounding the Northern, Central and Southern drill centres; and 11 locations surrounding the potential location of one more northerly and one more southerly drill centre. 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 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 2004 EEM Program. For sediment, no project-related effects were identified for metals other than barium. There was evidence that concentrations of hydrocarbons and barium were elevated by drilling activity near the Northern and Southern drill centres, and equivocal evidence that fines and sulphur concentrations may also have been elevated near these drill centres. No contamination was noted at the Central drill centre, where

drilling had been limited. 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 at the Northern and Southern drill centres did not extend beyond the 8 km zone of influence predicted by drill cuttings modeling (Hodgins and Hodgins 2000). Hydrocarbon contamination extended to between 5 and 8 km from source. Barium contamination extended to approximately 2 km from source. Any contamination from fines and sulphur was limited to within 1 km from source.

Directional effects were noted for both hydrocarbon and barium contamination in 2004, with dispersion primarily to the southeast. This is consistent with current records at White Rose for 2003 and 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.

Overall, there was little evidence of effects on benthic invertebrate communities. However, total abundance and the relative abundance of amphipods may have been affected by drilling. In 2004, total abundance and the relative abundance of amphipods were lower near the Southern drill centre. This pattern was not observed in the 2000 Baseline sampling program. The relative abundance of amphipods also decreased with increasing concentrations of hydrocarbons.

For both total abundance and the relative abundance of amphipods, decreases were mostly a function of the absence of high numbers, and not the occurrence of unusually low numbers, near the Southern drill centre. At stations greater than 2 km from the drill centre, both high and low numbers occurred for total abundance and the relative abundance of amphipods.

The apparent zone of effects on total abundance and the relative abundance of amphipods extended beyond the 500-m zone of effects predicted in the White Rose EIS. Nevertheless, White Rose results appear to be generally consistent with the recent literature on effects of contamination from offshore oil developments.

Additional sampling will be required at White Rose, as part of the scheduled 2005 EEM program, to determine if the spatial patterns in benthic invertebrate communities observed in 2004 are sustained and thus potentially project-related or if they represent natural year-to-year variability.

Biological 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 the more remote Reference Areas. Results for both plaice and crab are consistent with EIS predictions.

Conclusion

Overall, project-effects at White Rose in 2004 were limited. The spatial extent and magnitude of sediment contamination were within the ranges predicted in the EIS. If effects on benthos occurred, the spatial extent of this response exceeded predictions made in the EIS but was consistent with the recent literature on effects at other offshore oil developments. Sediment contamination and possible 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.

Acknowledgements

The White Rose Program (2004) 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 Craig Hollett, Barry Wicks, Matthew Hynes, Robbie Coish, Roy Skanes and Darroch Taylor. Fugro Jacques Geosurvey's Inc. provided geopositional 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 PSC 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. Taste tests results were interpreted by Dr. Joe Kiceniuk. 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, Theresa Fry and Paula Dalton. Dr. Malcolm Stephenson (Jacques Whitford) reviewed the document before final printing.

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1.0 Introduction

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).

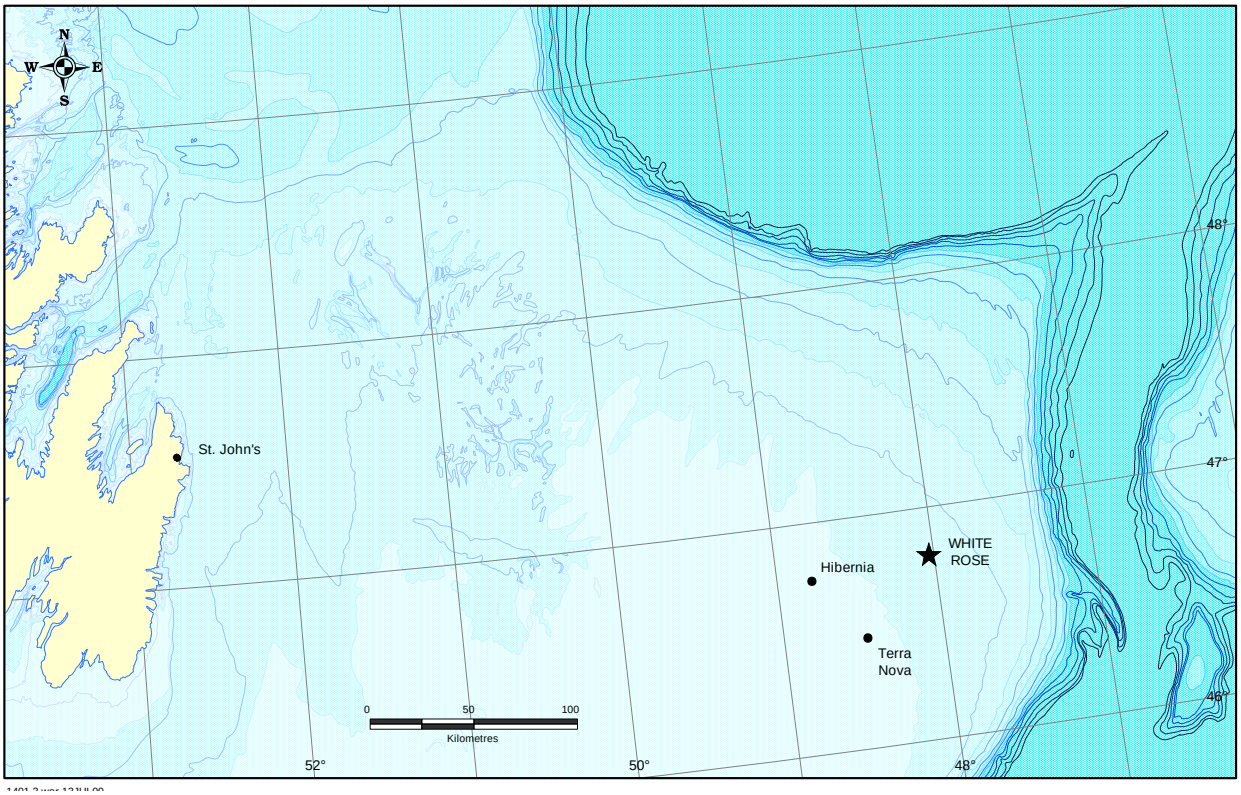


Figure 1-1 Location of the White Rose Oilfield

To date, development wells have been drilled at three drill centres: the Northern, Central and Southern drill centres. Drilling may also occur at two additional centres, one to the north of current centres (NN drill centre) and one to the south of current centres (SS drill centre) (Figure 1-2).

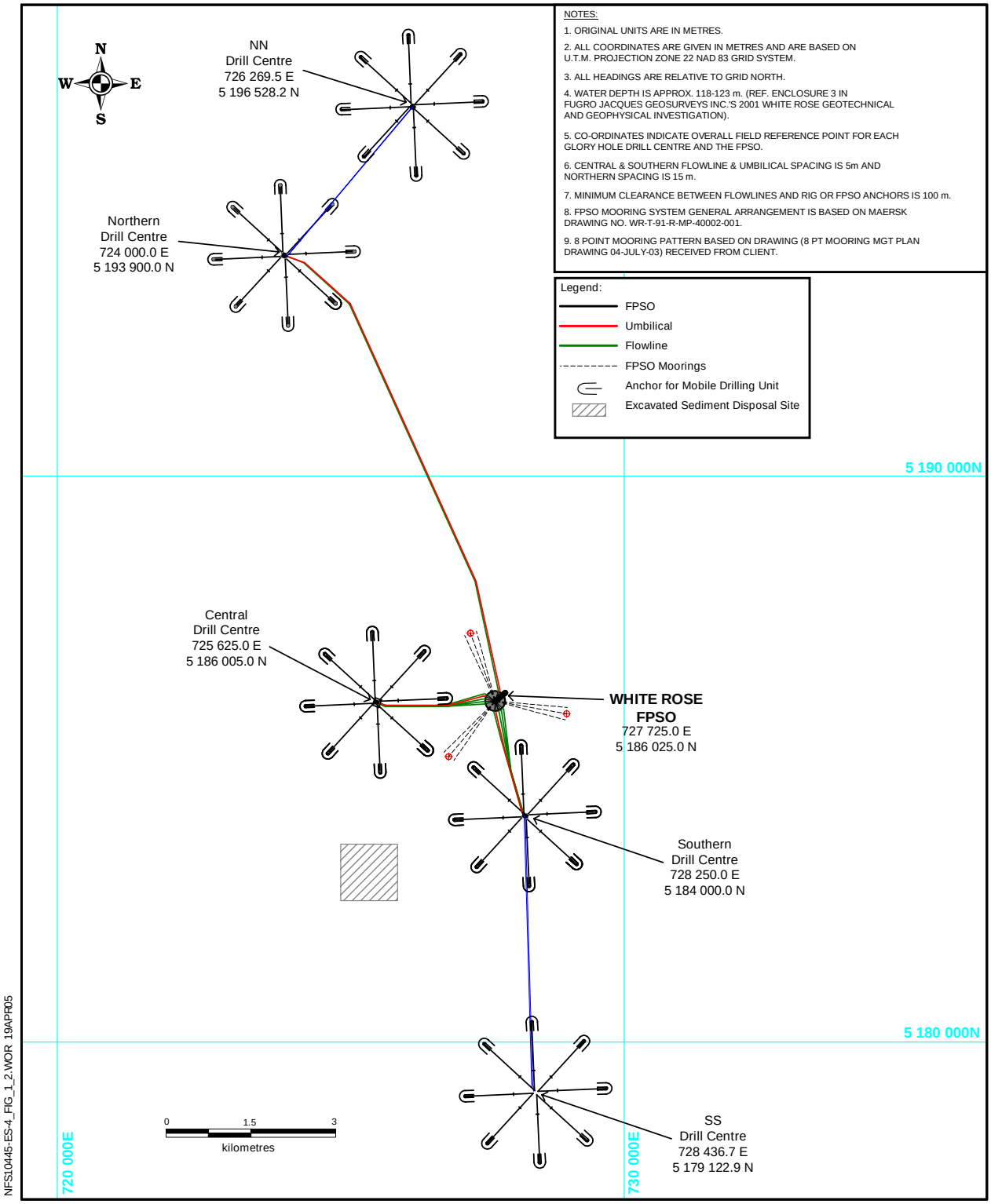


Figure 1-2 White Rose Field Layout

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 that in correspondence to the White Rose Public Hearings Commissioner, Husky Energy stated its intent to make both EEM reports and environmental compliance monitoring information "publicly available to interested stakeholders in a timely manner".

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 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) will continue to provide input on interpretation of EEM results and on program refinements, as required. WRAG comments on the 2004 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

EIS predictions (Husky Oil 2000) on physical and chemical characteristics of sediment and water, and predictions on benthos, fish and fisheries apply to the Husky Energy EEM program.

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 expect to have the greatest effect on physical and chemical characteristics of water, through release of produced water. The zone of influence for these two waste streams, defined here as the zone where project-related physical and chemical alterations might occur, was not expected to extend beyond approximately 8 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 to be mild within approximately 500 m of drill centres but fairly large in the immediate vicinity 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 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) 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 2004 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 Design document (Husky Energy 2004).

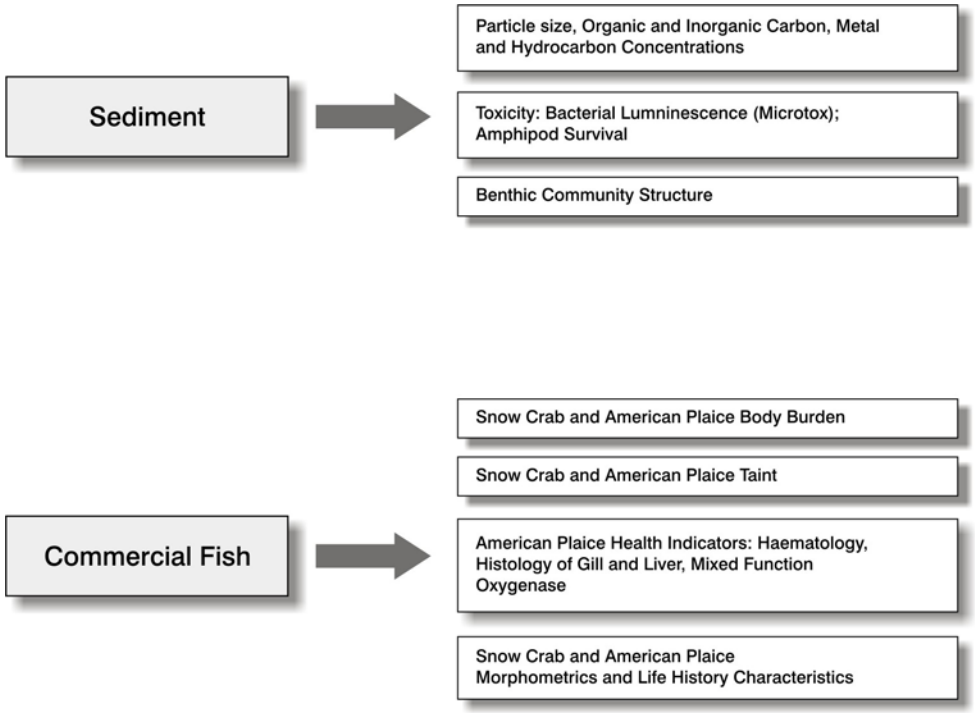


Figure 1-3 EEM Program Components

Note: modified from Petro-Canada 2003

1.7 Monitoring Hypotheses

Monitoring, or null (H_0), 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 (H_0) 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:
  - H_0: There will be no change in SQT variables with distance or direction from project discharge sources over time.
- Commercial Fish:
  - $H_0(1)$: 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 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 both the Baseline Characterization ("baseline") and EEM program, 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, or 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 and 2004 EEM program. A total of 48 sediment stations were sampled during baseline and 56 stations were sampled for EEM program; 37 stations were common to both 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. 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 EEM program 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 other stations (see Husky Energy 2004 for details).

Station additions for the EEM program include four new 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 new drill centre stations located approximately 300 m from each of the Northern, Central and Southern drill centres, and six new drill centre stations located 1 km from the proposed location of each of the SS and NN drill centres. As was the case for drill centre stations around the Northern, Central and Southern drill centres, some stations around the SS and NN drill centres will be deleted in future EEM programs once the locations of these drill centres becomes known. Table 1-1 provides a summary of these changes as well as stations name changes that were proposed in the EEM design document to simplify reporting of results. Figure 1-4 and 1-5 show baseline and EEM station locations.

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 (see Figure 1.5). 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).

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
Deleted	F1-18,000	Deleted	GH3-1
Deleted	F5-18,000	Deleted	GH3-2
Deleted	SS and NW Reference	Deleted	GH3-4

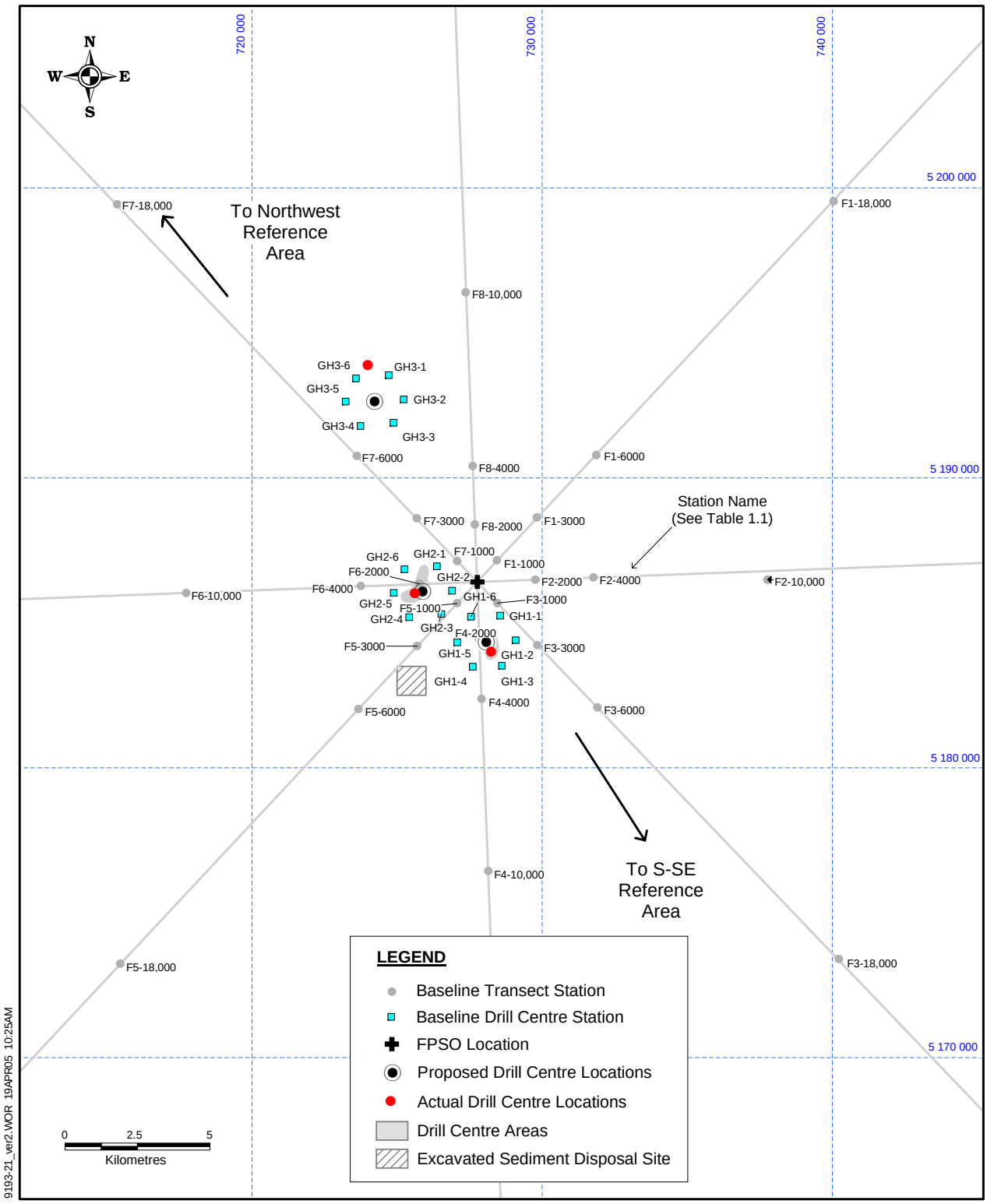


Figure 1-4 Baseline Program Survey Design

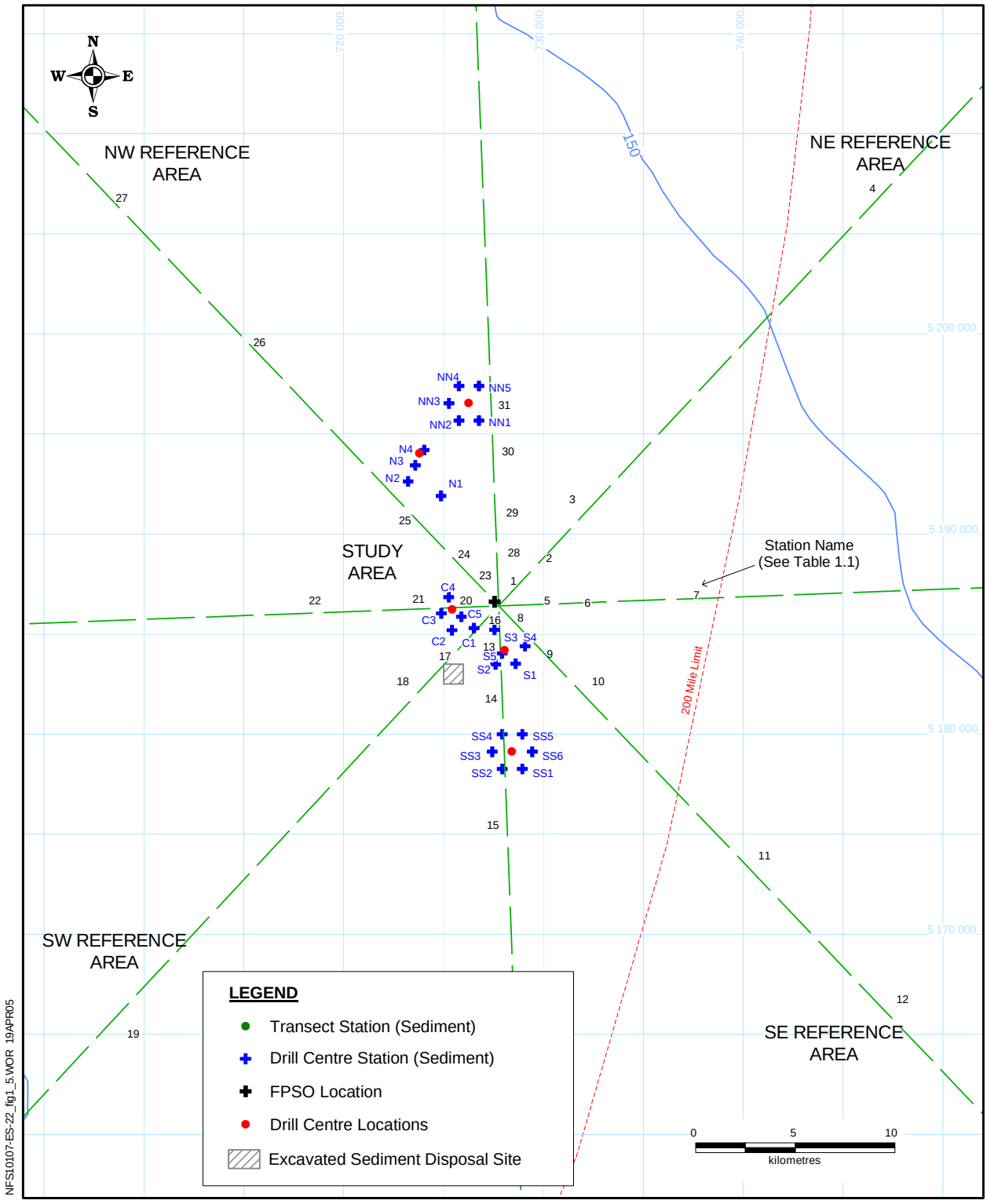


Figure 1-5 EEM Program Survey Design

2.0 Scope

This document, *White Rose Environmental Effects Monitoring Program 2004 (Volume 1)*, provides summary results, analysis and interpretation for the White Rose 2004 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 program are compared to 2004 results. Since analysis results are often highly technical, a key 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 2004.

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 2004 (Volume 2))*. Raw data and other information supporting *Volume 1* are also provided in *Volume 2*.

3.0 Acronyms

The following acronyms are used in this report.

ANOVA	Analysis of Variance
BC	Bray-Curtis (measure of similarity)
BTEX	Benzene, Toluene, Ethylbenzene and Xylene
CCME	Canadian Council of Ministers of the Environment
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
FPSO	Floating Production, Storage and Offloading (facility)
HC	Hydrocarbon
ISQG	Interim Sediment Quality Guidelines
MFO	Mixed Function Oxygenase
MSDS	Material Safety Data Sheet
MDS	Multidimensional Score
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
SQT	Sediment Quality Triad
SR	Study versus Reference
TEL	Threshold Effects Levels
TIC	Total Inorganic Carbon
TOC	Total Organic Carbon
TPH	Total Petroleum Hydrocarbon
UCM	Unresolved Complex Mixture
WBM	Water-Based Mud

4.0 Project-Related Activities and Ocean Currents

4.1 Introduction

This section reports on construction, installation and drilling activities in the White Rose field. The section also summarizes the discharges and spills associated with these operations from October 2003 through October 2004 and provides information on surface, mid-water and bottom currents at White Rose over this time period.

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 three general categories:

- construction and installation activities;
- supply vessel operations; and
- drilling operations.

In late 2005, producing operations (i.e., oil and gas production, storage and offloading to a tanker) will commence at the White Rose Field once hook up, commissioning and introduction of hydrocarbons to the FPSO *SeaRose* have been completed. By that time, all construction and installation activities will also have been completed, leaving ongoing development and delineation drilling, supply and production operations to continue. Producing operations will continue for an estimated 15 years while drilling operations are expected to be complete after five to seven years.

4.2.1 Construction and Installation Operations

Construction and installation activities started in the summer of 2002 and have continued through to 2004. 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 will be mated in the third or fourth quarter of 2005. The remainder of the flowlines will be installed in 2005. These and flowlines previously laid will then be connected to the FPSO.

The largest physical disturbance to the sea floor to date has been the excavation of the glory holes at the three drill centres. A total of approximately 356,000 m^3 of seabed material, predominately sand with gravel (>95%) 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 (Figure 1-2, Section 1).

During the construction and installation activities that took place between 2002 and October 2003, less than 100 L of hydraulic fluid was spilled from all vessel and equipment sources.

Losses during the October 2003 to October 2004 period are summarized in Table 4-1.

Table 4-1 Summary of Environmental Losses from White Rose Offshore Operations - October 2003 to October 2004

Operation	Hydrocarbons	Drilling Fluids	Other
Drilling	1 liter of hydraulic fluid lost from crane in one incident	99.1 cubic meters of synthetic drilling fluid lost in two incidents	-
ROV Operations - Drilling	77 liters of hydraulic fluid lost during ROV operations in ten incidents	-	-
ROV Operations – Construction	32 liters of hydraulic fluid lost during ROV operations in three incidents	-	-
Well Testing	115 liters of crude oil lost during well testing in seven incidents	-	-
Supply Vessel Operations	20 liters of hydraulic fluid lost from thrusters in one incident	-	apparent evidence of night collision with marine mammal in one incident loss of 5 empty containers overboard in transit to port in one incident
Construction Vessel Operations	15 liters of hydraulic fluid lost in one incident	-	-

Note: - ROV = Remotely Operated Vehicle; SBM = Synthetic-Based Mud

4.2.2 Supply Vessel Operations

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. Losses from vessel operations other than these authorized waste streams during the October 2003 to October 2004 are summarized in Table 4-1.

4.2.3 Drilling Operations

4.2.3.1 Drilling Discharges

Husky Energy employs both water-based muds (WBMs) 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.

Apart from direct drilling discharges, there is a need to remove accumulated cuttings and surplus concrete from the drill centres. This is accomplished with an Remotely Operated Vehicle (ROV) equipped with cutter suction and discharge equipment that removes the excess material and discharges it approximately 50 m from the edge of the drill centre through a diffuser. This activity occurred predominantly at the Southern drill centre from January to May 2004 (approximately 900 hours). In September and October, approximately 500 hours were invested in this work at the Central drill centre and 145 hours were spent at this activity at the Northern drill centre in April.

Water-Based Drilling Discharges
From October 2003 to October 2004, the total mass of drill cuttings and WBMs discharged to the sea floor at the three drill sites on the White Rose field was 20,279 metric tonnes of which 18,610 metric tonnes were rock cuttings and 1,670 metric tonnes were WBMs.

These WBMs and cuttings discharges occurred at the three drill sites in the following proportions: 68% at the Southern drill centre (9 upper well sections), 15% at the Northern drill centre (2 upper well sections) and 17% at the Central drill centre (2 upper well sections).

Synthetic-Fluid-Based Drilling Discharges
From October 2003 to October 2004, the total mass of drill cuttings and SBM-on-cuttings discharged to the sea floor at the three drill sites on the White Rose field was 5434 metric tonnes of which 5434 metric tonnes were rock cuttings and 478 metric tonnes were SBM-on-cuttings.

These SBM and cuttings discharges occurred at the three drill sites in the following proportions: 91% at the Southern drill centre and 9% at the Northern drill centre. No SBM drilling occurred at the Central drill centre during the reporting period.

The C-NLOPB's Offshore Waste Treatment Guidelines require operators to target a value of 6.9% or less SBM-on-cuttings. Depending on drilling conditions in different wells and well section performance, the 6.9% target has varied from approximately 3.8% to 13.3% SBM-on-cuttings based on 48 hour rolling averages.

Table 4-2 shows Husky Energy's performance with regard to the 6.9% target by drill centre.

Table 4-2 % Synthetic Oil on Cuttings for Well Sections Drilled with SBM

Drilling Site	Southern Drill Centre	Northern Drill Centre	Central Drill Centre
Range of SBM-on-cuttings for each well drilled	6.73 to 10.76 % 3.84 to 12.60 % 6.21 to 11.51 % 7.75 to- 10.15 %	4.41 to 13.32 %	No SBM drilling was carried out during reporting period

Completion Fluids

On completion, the well bore needs to be cleaned of residual cuttings. This is done by flushing with completions fluids consisting mainly of brine. During the reporting period, approximately 43 m^3 of completion fluids were discharged from wells at the Northern drill centre, and approximately 225 m^3 were discharged from wells at the Southern drill centre. No completion operations were carried out at the Central drill centre during the reporting period.

4.2.3.2 Other Operational Discharges

The operational discharges from Husky Energy's drilling platform operations other than drill cuttings and drilling mud over the past year are summarized below in Table 4-3.

Table 4-3 Operational Discharges from 2003 to 2004

Authorized Discharges (m^3)	2003			2004									
	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct
Bilge Water[1]	14	2	0	11.1	3	0	4.5	12.5	5.5	9	0	10.8	11
Glycol-based fluid from BOP[2]	0	6.6	5.7	7	7.6	3.9	0.5	2.1	0.7	0.9	6.9	2.7	10.7
Deck Drainage[3]	105.9	100.5	201.4	106.5	121.3	193	119	80	115	167.5	143	87	208
Glycol-based fluid from Subsea Equipment[4]	0	0	0	0	0	0	0	168	129	0	0	0	0

Notes:
- [1] bilge discharges are maintained at 15 ppm or less
- [2] BOP (Blow-Out-Preventor) testing is to ensure functionality and therefore safety and environmental protection; volumes are the amount of active ingredient i.e., glycol and erifon at maximum of 42 and 2% respectively of total volume discharged
- [3] deck drainage discharges are maintained at 15 ppm or less
- [4] losses from subsea equipment during hookup and installation work during is unavoidable; volumes are the amount of active ingredient i.e., glycol and triethanolamine at a maximum of 70% and of 5% of total volume discharged

4.3 Ocean Currents

Current direction and speed from oceanographic equipment moored at White Rose over the reporting period are provided in Table 4-4 and in current roses[a] displayed in Figures 4-1 to 4-15. Currents to the south have been common at all depths. Average current speeds at the surface, mid-depth and bottom from the last quarter of 2003 to the last quarter of 2004 were 17 cm/sec, 14 cm/sec and 15 cm/sec, respectively. The maximum current speed recorded was 81 cm/sec, in the last quarter of 2004, at the surface.

Table 4-4 Current Direction and Speed in 2003 and 2004

Depth	Time Interval	Predominant Direction	Mean Speed (cm/s)	Maximum Speed (cm/s)
Surface (23 to 27 m)	Q4 2003	South	14.83	63
	Q1 2004	South	17.76	51
	Q2 2004	East	14.05	44
	Q3 2004	South	14.87	61
	Q4 2004	South	23.72	81
Mid-Depth (55 to 59 m)	Q4 2003	Southwest	12.97	42
	Q1 2004	Southwest	13.31	39
	Q2 2004	Southeast	13.18	45
	Q3 2004	South-Southwest	12.77	62
	Q4 2004	South-Southeast	17.83	75
Bottom (95 to 99 m)	Q4 2003	South	11.84	37
	Q1 2004	South	14.40	37
	Q2 2004	South	14.76	51
	Q3 2004	South	15.71	72
	Q4 2004	South-Southeast	17.29	65

Note: - Observations may not have been collected over the entire time interval. Refer to Figures 4-1 to 4-15 for observation periods.

[a] Current Rose Description

A current rose illustrates the percent frequency of distribution of current direction and speed for a given time period and at a given depth, e.g., October to December, at 23 m. The tabular listing on the right side reports the number of observations and total percentage for each of eight compass directions, as well as any calm or missing values. The current rose on the left presents this same directional frequency as well as the distribution of current speed within each directional sector or bar. Bars represent the percentage frequency of current observed to each direction. Each circle equals 5%. Each section of a current rose bar corresponds to currents of a given speed range or bin, with bins being the noted 5 cm/s in size. The section length (radial distance out from the middle of the rose) is the percentage of all observations that are in a given speed range, for the given direction. The number reported in the inner circle represents the percentage of calm observations. The section widths increase in size as the speed range increases and as the bar extends out from the origin. The first bar section is a line segment (0 width), and each subsequent bar is a rectangle. The length of the first bar section represents the percentage of observations in the speed range 0 to 5 cm/s, the length of the second bar section represents the percentage of observations in the speed range 5 to 10 cm/s, and so on. A bar with 14 sections will therefore report a percentage of observations up to its largest section or speed range of 55 to 60 cm/s.

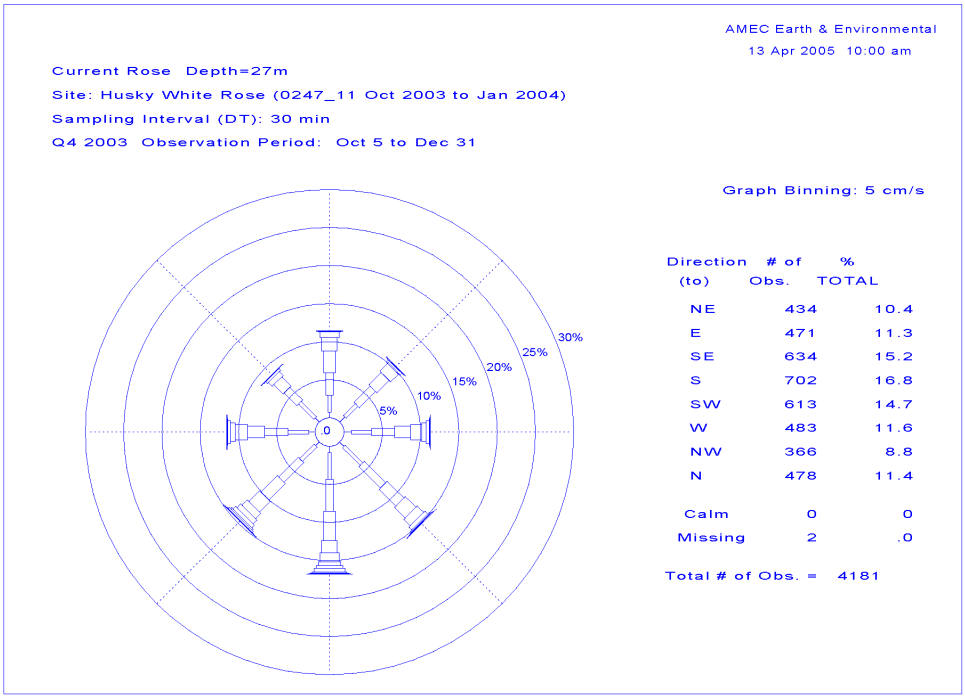


Figure 4-1 Surface Currents, Q4 2003

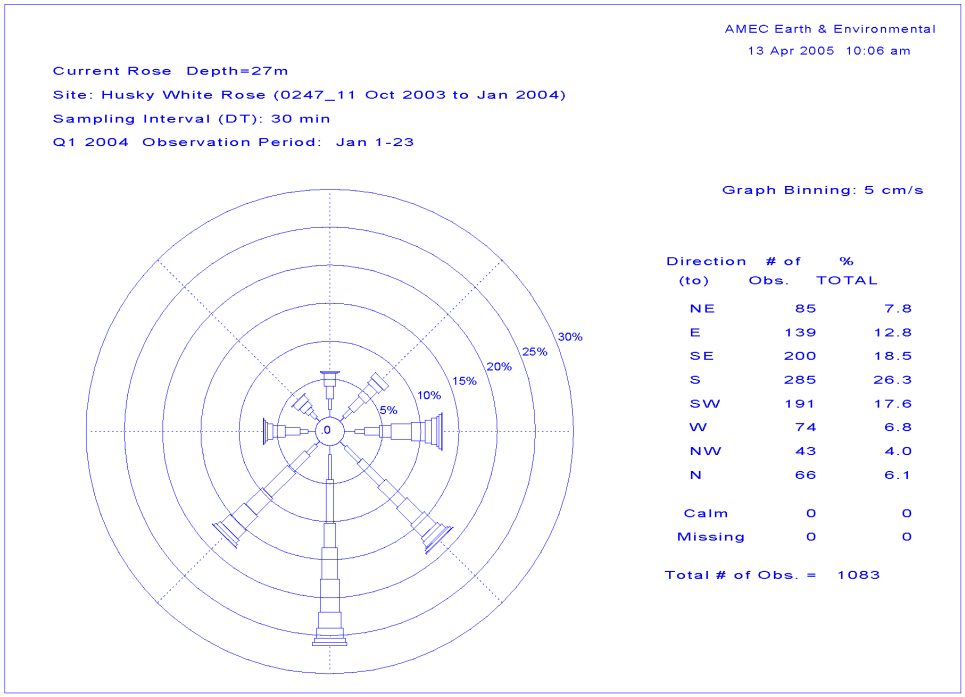


Figure 4-2 Surface Currents, Q1 2004

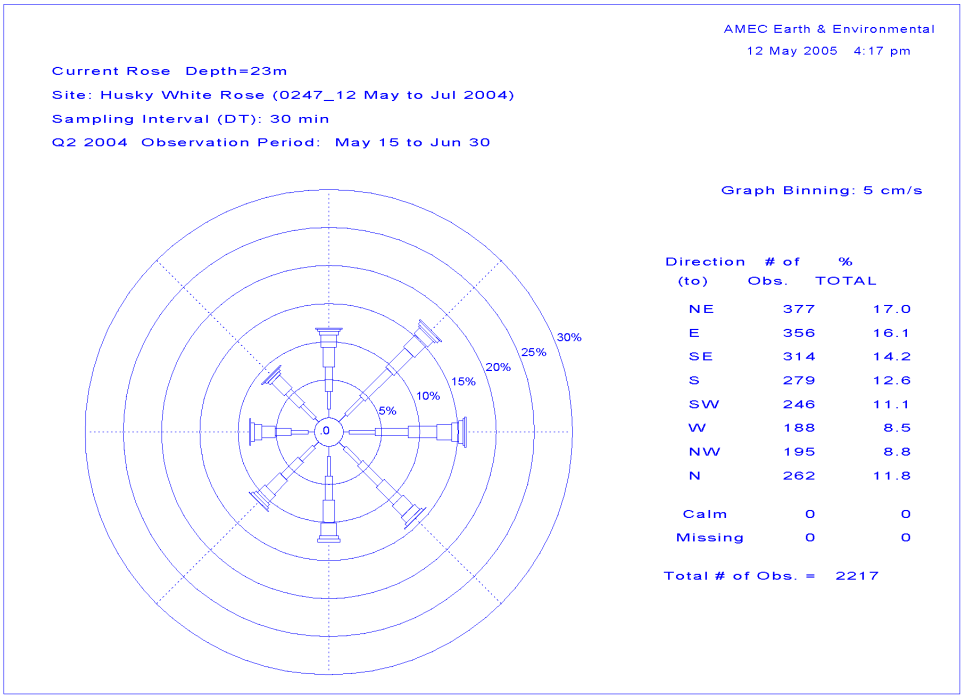


Figure 4-3 Surface Currents, Q2 2004

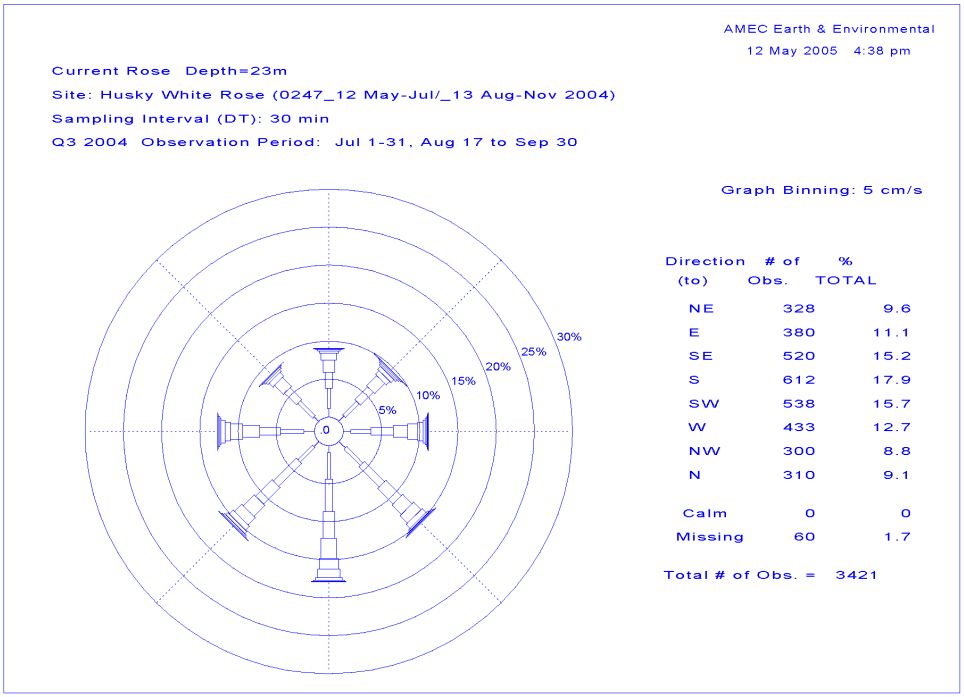


Figure 4-4 Surface Currents, Q3 2004

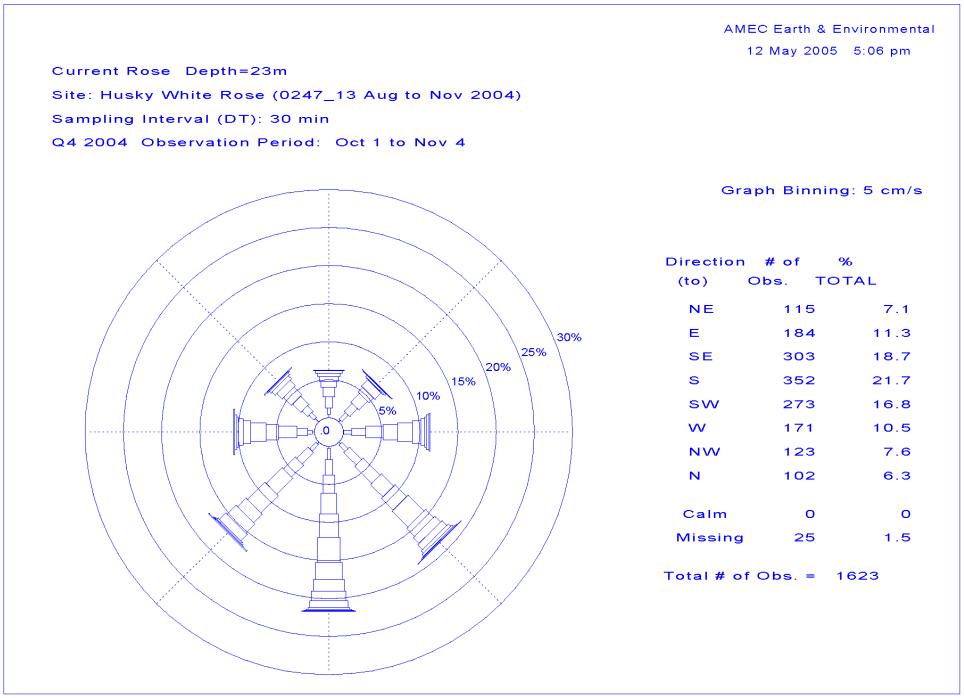


Figure 4-5 Surface Currents, Q4 2004

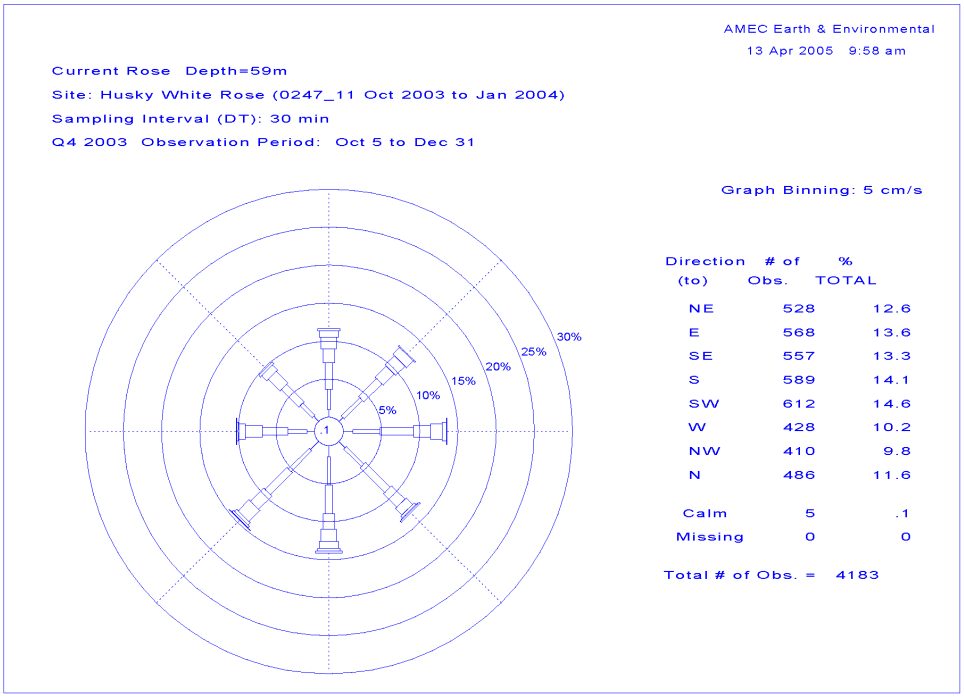


Figure 4-6 Mid-Depth Currents, Q4 2003

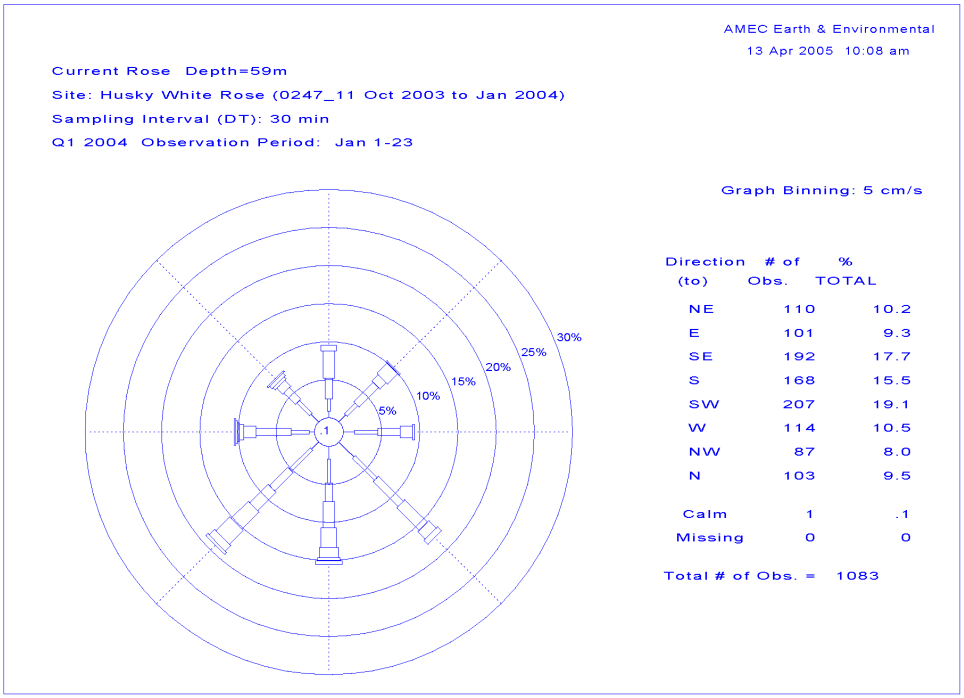


Figure 4-7 Mid-Depth Currents, Q1 2004

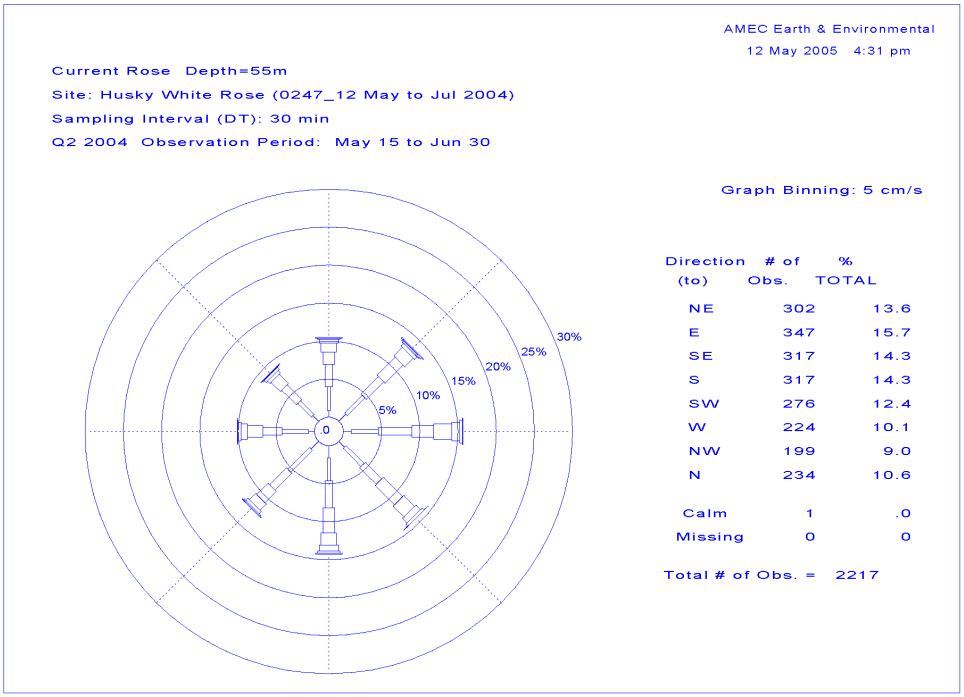


Figure 4-8 Mid-Depth Currents, Q2 2004

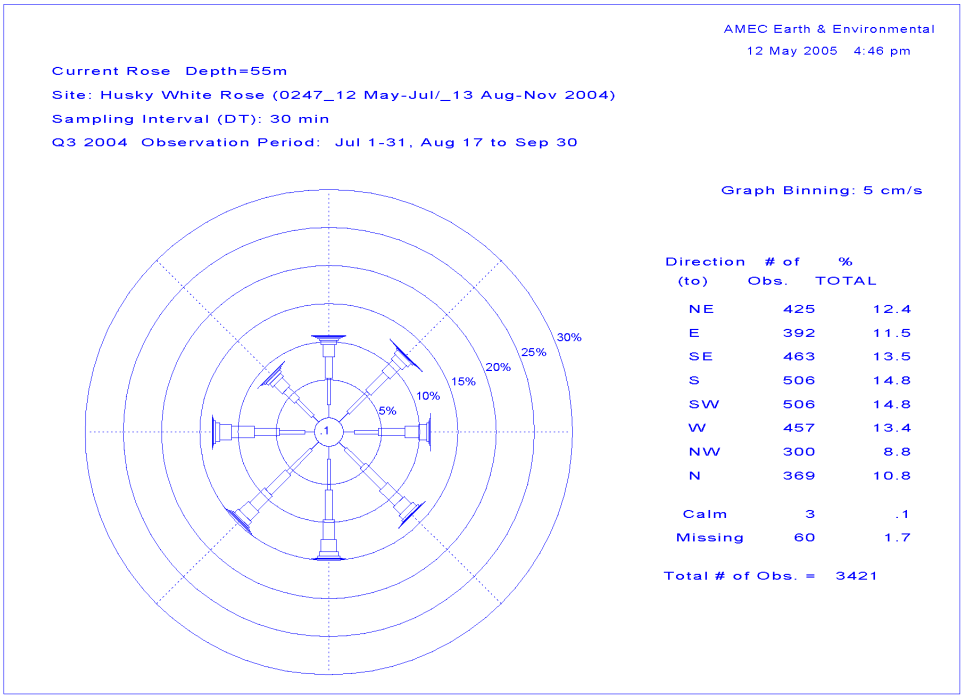


Figure 4-9 Mid-Depth Currents, Q3 2004

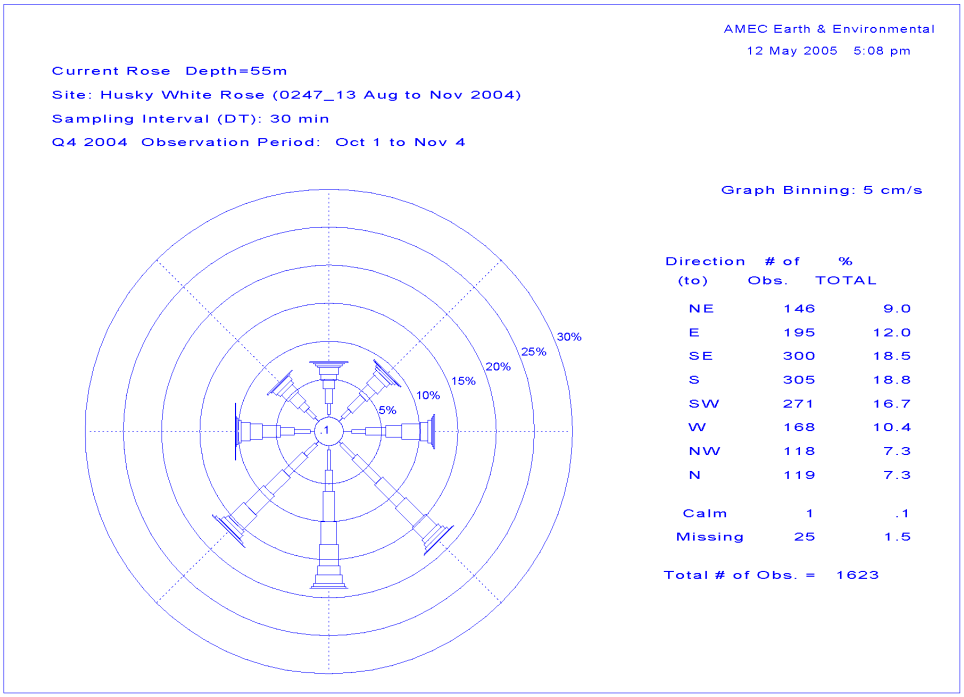


Figure 4-10 Mid-Depth Currents, Q4 2004

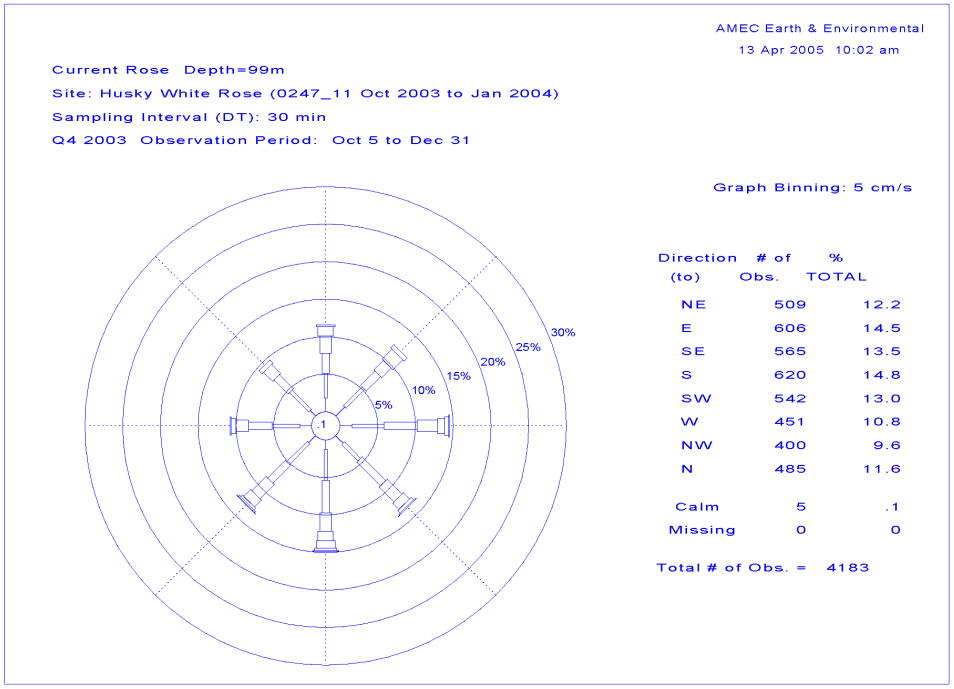


Figure 4-11 Bottom Currents, Q4 2003

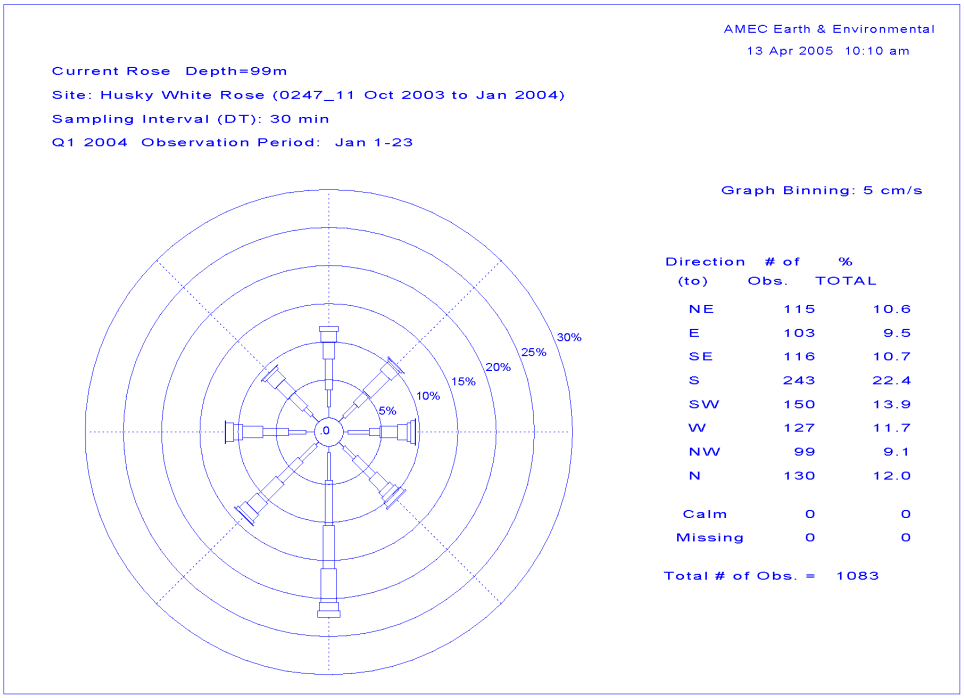


Figure 4-12 Bottom Currents, Q1 2004

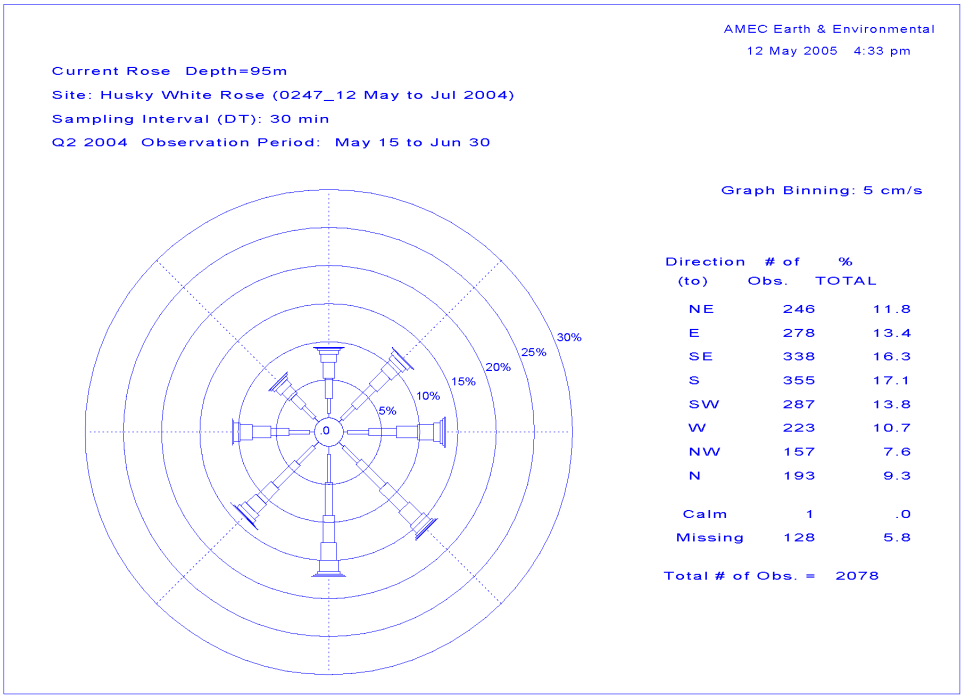


Figure 4-13 Bottom Currents, Q2 2004

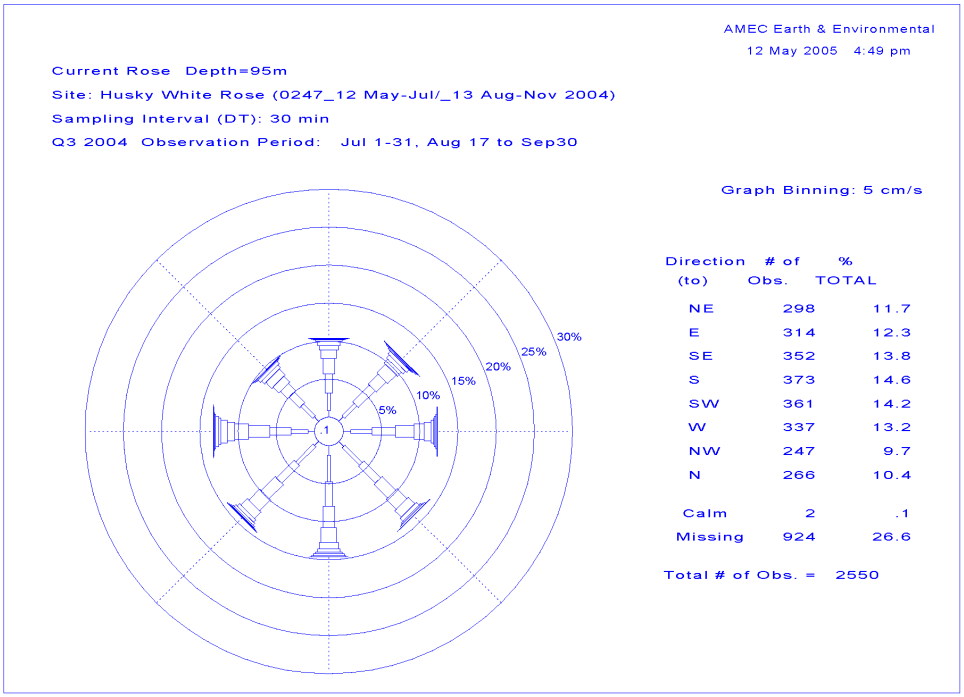


Figure 4-14 Bottom Currents, Q3 2004

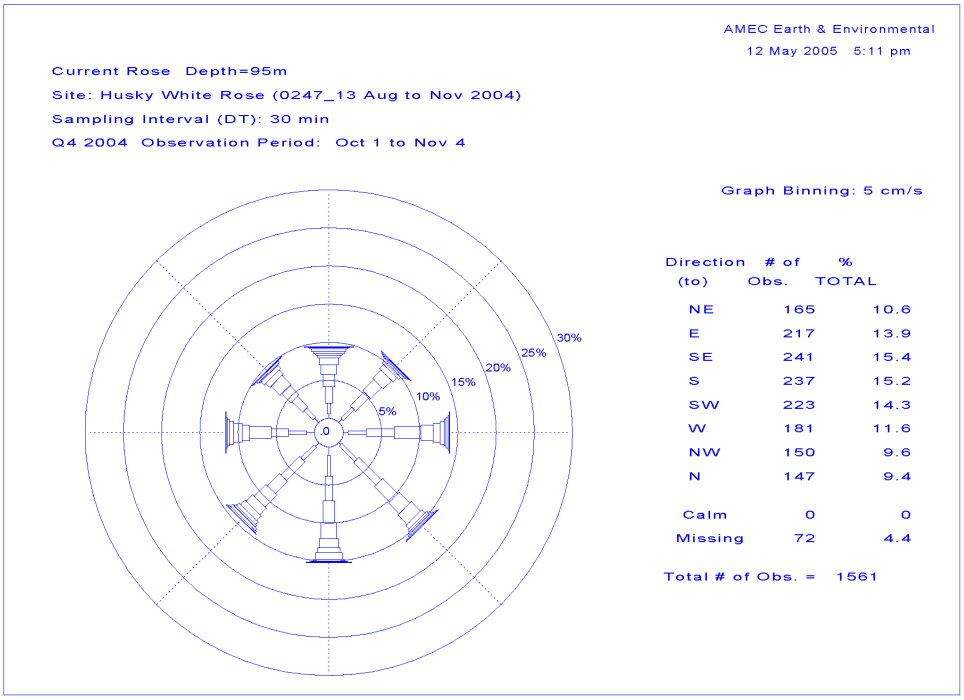


Figure 4-15 Bottom Currents, Q4 2004

5.0 Sediment Component

5.1 Field Collection

The sediment component of the 2004 EEM Program was conducted from September 26 to October 11, 2004 using the offshore supply vessel *Burin Sea*. Sampling dates for the baseline program and the 2004 EEM program are shown in Table 5-1. Sediment stations for the baseline and 2004 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). Geographic coordinates and distances to drill centres for EEM stations are provided in Appendix B-1.

Table 5-1 Dates of Previous Field Programs

Trip	Date
Baseline Program	September 9 to September 19, 2000
2004 EEM Program	September 26 to October 11, 2004

Sediment samples were collected using a large-volume box corer (mouth diameter = 35.6 cm, depth = 61 cm) designed to mechanically take an undisturbed sediment sample over approximately 0.1 m^2 of seabed (Figures 5-1 and 5-2). Three boxcores 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 the top of all three boxcores (Figure 5-3). These were stored in pre-labelled 250-mL glass jars at -20°C. Sediment samples collected for toxicity were collected from the top 7.5 cm of one boxcore and stored at in the dark at 4°C in a 4-L high-density food-grade polyethylene bucket with an O-ring seal (amphipod toxicity) and a sterile 200 ml Whirl-Pak (bacterial luminescence). Sediment samples for benthic community structure analysis were collected from the top 15 cm of two boxcores and stored in two separate 11-L pails. These samples were preserved with approximately 1 L of 10% buffered formalin.

Sediment chemistry field blanks composed of clean sediment obtained from PSC Maxxam Analytics were collected for stations 29, C4 and SS5. Blank vials were opened as soon as 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 27, 31, NN1, S4, SS2 and SS4. Both field blanks and field duplicates were assigned randomly to stations.

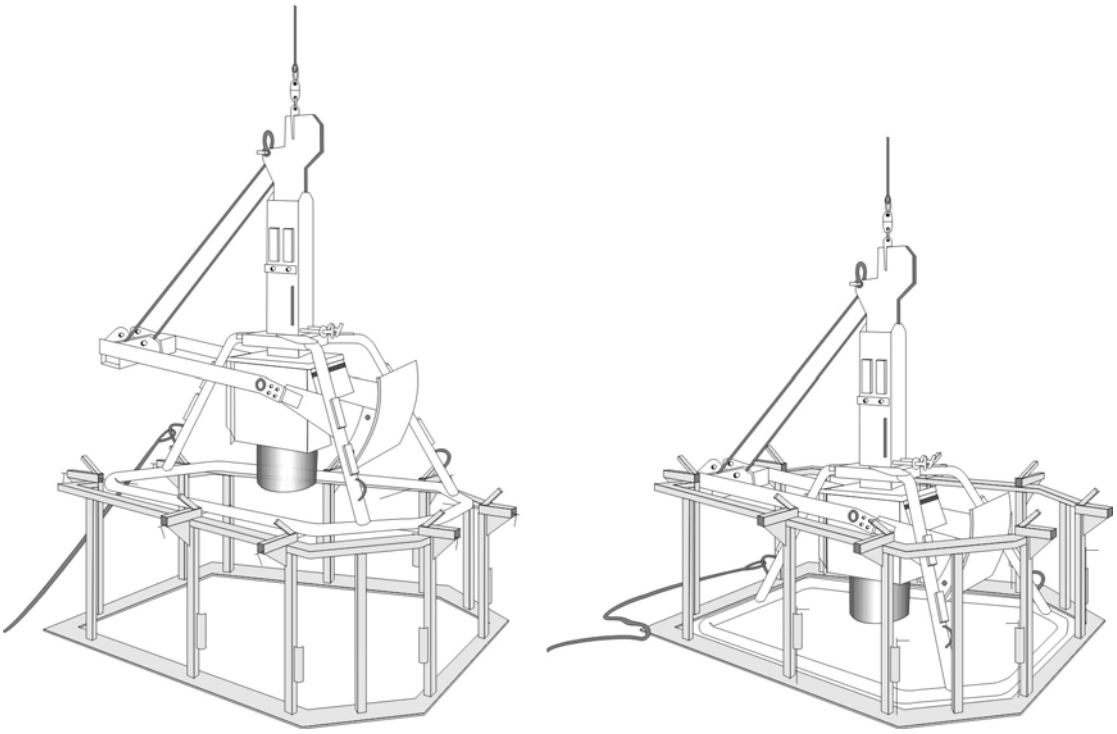


Figure 5-1 Box Corer Diagram

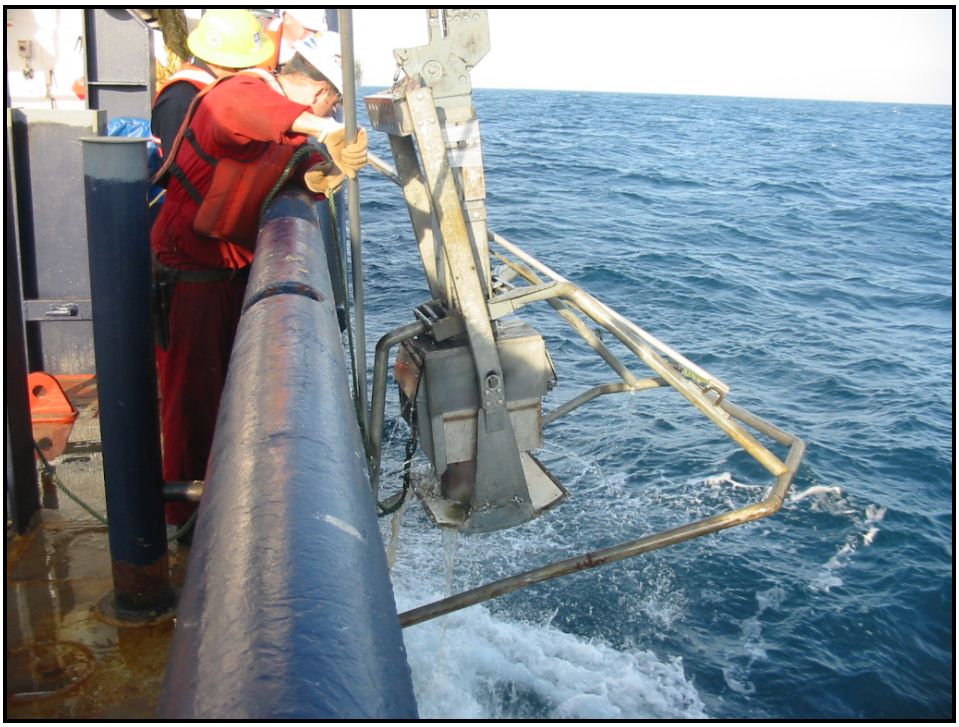


Figure 5-2 Box Corer

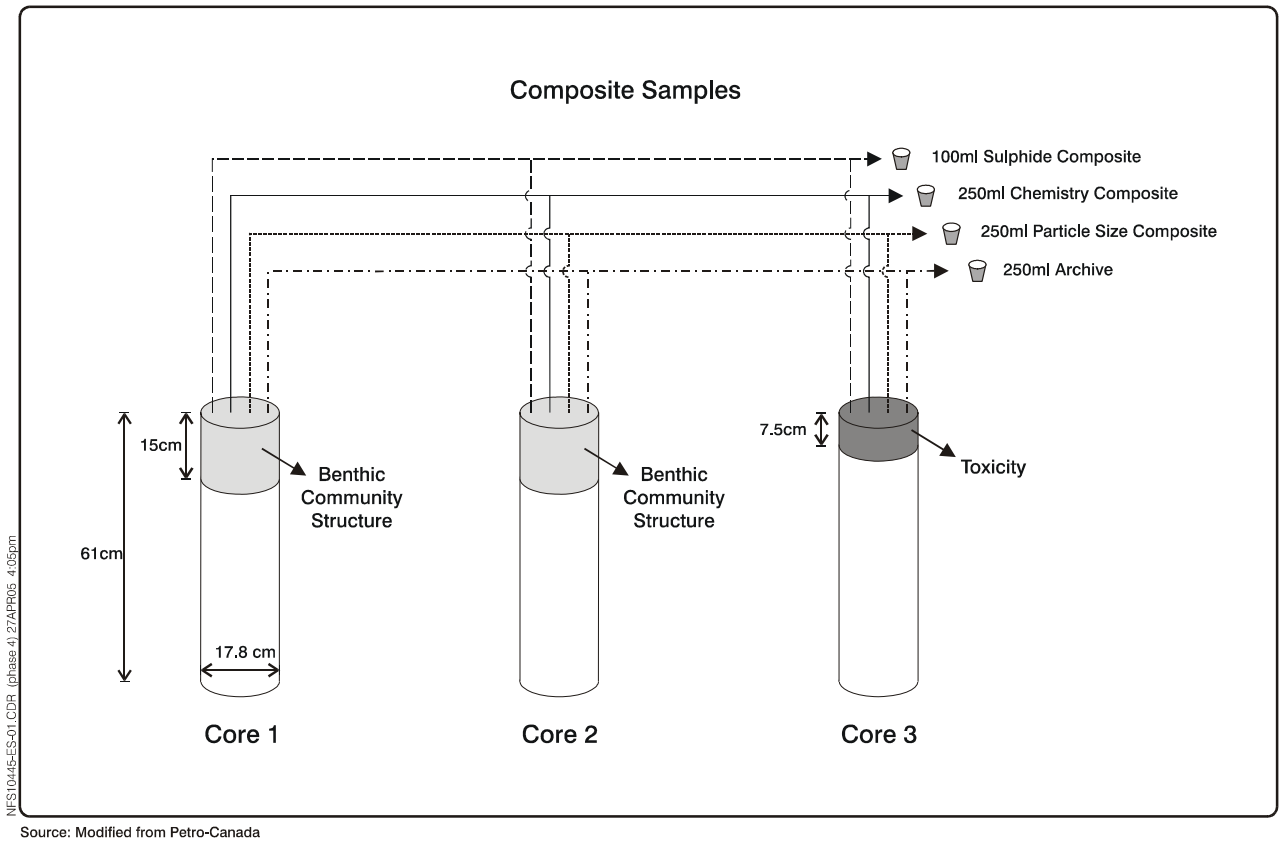


Figure 5-3 Allocation of Samples from Cores

Standard 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 PSC 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 and 2004)

Variables	Method	2000 EQL	2004 EQL	Units
HCs				
Benzene	Calculated	0.025	0.025	mg/kg
Toluene	Calculated	0.025	0.025	mg/kg
Ethylbenzene	Calculated	0.025	0.025	mg/kg
Xylenes	Calculated	0.05	0.05	mg/kg
C_6-C_{10}	Calculated	2.5	2.5	mg/kg
>C_{10}-C_{21}	GC/FID	0.25	0.25	mg/kg
>C_{21}-C_{32}	GC/FID	0.25	0.25	mg/kg
>C_{10}-C_{32}	Calculated	0.5	0.5	mg/kg
C_6-C_{32} (TPH)	Calculated	3	3.2	mg/kg
PAHs				
1-Chloronaphthalene	GC/FID	NA	0.05	mg/kg
2-Chloronaphthalene	GC/FID	NA	0.05	mg/kg
1-Methylnaphthalene	GC/FID	0.05	0.05	mg/kg
2-Methylnaphthalene	GC/FID	0.05	0.05	mg/kg
Acenaphthene	GC/FID	0.05	0.05	mg/kg
Acenaphthylene	GC/FID	0.05	0.05	mg/kg
Anthracene	GC/FID	0.05	0.05	mg/kg
Benz[*a*]anthracene	GC/FID	0.05	0.05	mg/kg
Benzo[*a*]pyrene	GC/FID	0.05	0.05	mg/kg
Benzo[*b*]fluoranthene	GC/FID	0.05	0.05	mg/kg
Benzo[*ghi*]perylene	GC/FID	0.05	0.05	mg/kg
Benzo[*k*]fluoranthene	GC/FID	0.05	0.05	mg/kg
Chrysene	GC/FID	0.05	0.05	mg/kg
Dibenz[*a,h*]anthracene	GC/FID	0.05	0.05	mg/kg
Fluoranthene	GC/FID	0.05	0.05	mg/kg
Fluorene	GC/FID	0.05	0.05	mg/kg
Indeno[1,2,3-*cd*]pyrene	GC/FID	0.05	0.05	mg/kg
Naphthalene	GC/FID	0.05	0.05	mg/kg
Perylene	GC/FID	0.05	0.05	mg/kg
Phenanthrene	GC/FID	0.05	0.05	mg/kg
Pyrene	GC/FID	0.05	0.05	mg/kg
Carbon				
Total Carbon	LECO	0.1	0.2	g/kg
Total Organic Carbon	LECO	0.1	0.2	g/kg
Total Inorganic Carbon	By Diff	0.2	0.3	g/kg
Metals				
Aluminum	ICP-MS	10	10	mg/kg
Antimony	ICP-MS	2	2	mg/kg
Arsenic	ICP-MS	2	2	mg/kg
Barium	ICP-MS	5	5	mg/kg
Beryllium	ICP-MS	5	2	mg/kg
Cadmium	GFAAS	0.05	0.05	mg/kg

Variables	Method	2000 EQL	2004 EQL	Units
Chromium	ICP-MS	2	2	mg/kg
Cobalt	ICP-MS	1	1	mg/kg
Copper	ICP-MS	2	2	mg/kg
Iron	ICP-MS	20	50	mg/kg
Lead	ICP-MS	0.5	0.5	mg/kg
Lithium	ICP-MS	5	2	mg/kg
Manganese	ICP-MS	2	2	mg/kg
Mercury	CVAA	0.01	0.01	mg/kg
Molybdenum	ICP-MS	2	2	mg/kg
Nickel	ICP-MS	2	2	mg/kg
Selenium	ICP-MS	2	2	mg/kg
Strontium	ICP-MS	5	5	mg/kg
Thallium	ICP-MS	0.1	0.1	mg/kg
Tin	ICP-MS	2	2	mg/kg
Uranium	ICP-MS	0.1	0.1	mg/kg
Vanadium	ICP-MS	2	2	mg/kg
Zinc	ICP-MS	2	5	mg/kg
Other				
Ammonia (as N)	COBAS	NA	0.25	mg/kg
Sulphide	SM4500	NA	2	mg/kg
Sulphur	LECO	NA	0.02	%(w)
Moisture	Grav.	0.1	0.1	%

Notes:
- The EQL is the lowest concentration that can be reliably achieved 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_6-C_{10} range commonly referred to as the gasoline range. >C_{10}-C_{21} is referred to as the diesel range and is the range where lightweight fuels like diesel will be detected. The >C_{21}-C_{32} range is where lubricating oils (i.e., motor oil and grease), crude oil, and in some cases, bunker C oil, would be detected. Total Petroleum Hydrocarbons (TPHs) encompass all three ranges (C_6-C_{32}). 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_6-C_{32} 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_{10}-C_{21} (MSDS for PureDrill IA-35 2000). Most of the components of PureDrill IA-35 form an UCM that starts around the retention time of C_{11} n-alkane (2.25 min) and ends around the same time as C_{21} 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_{17}-C_{18} size.

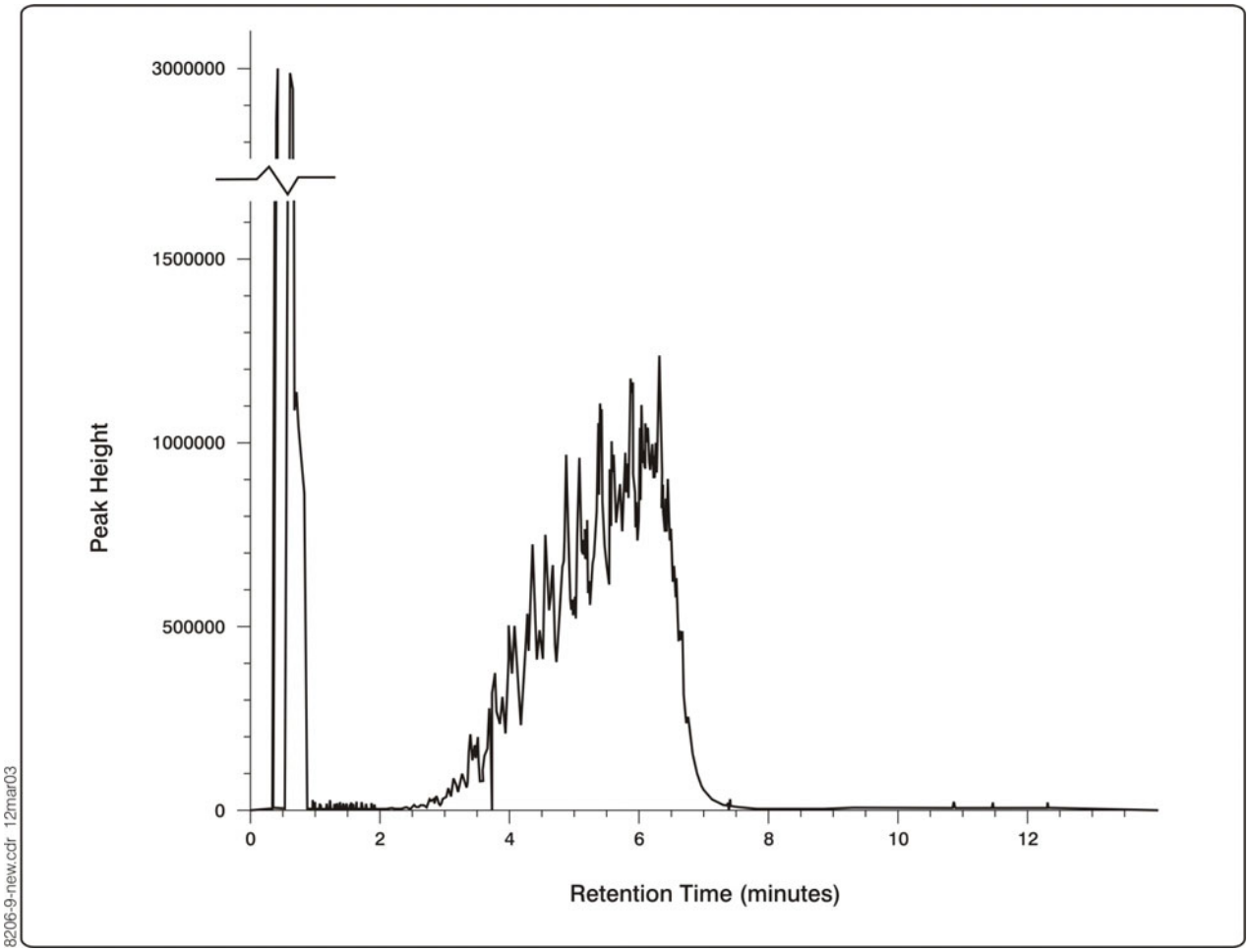


Figure 5-4 Gas Chromatogram Trace for PureDrill IA-35

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 four to five replicate 1-L test chambers (four replicates were used for some stations because of restricted amphipod availability) 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. A sixth 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.

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 (2002a) Reference Method 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 (2002a). Data from the 2000 (baseline) program were reexamined using the criteria outlined in Environment Canada (2002a) because analyses in 2000 were conducted using earlier Environment Canada guidelines (small volume solid phase assay; Environment Canada 1992a). Reinterpretation of 2000 data using Environmental Canada (2002a) did not alter any of the 2000 interpretations.

All toxicity tests were initiated within six weeks of sample collection, meeting the minimal requirements of sediment storage recommended by Environment Canada Guidelines (Environment Canada 1998; 2002a).

5.2.2.1 Results Interpretation

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 value. 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, calculated using TOXCALC computer program (Tidepool Scientific Software 1994).

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 is statistically significantly different than mortality in the negative control sediment.

For the bacterial luminescence assay, Environment Canada has published a new method reference method for Solid Phase Microtox™ Testing. The new reference method (Environment Canada EPS 1/RM/42 2002) contains new interim guidelines for assessing 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 and depth which is comprised of less than 20% fines and that has an IC50 of ≥ 1,000 mg/L, 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.

5.2.3 Benthic Community Structure

All 2004 samples were provided whole to Arenicola Marine Limited (Wolfville, Nova Scotia). 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.

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 from 2000 were processed by Pat Steward of Envirosphere Ltd. Methods and the level of taxonomy were similar to those used for the 2004 samples (see Husky Energy 2001 for details).

5.3 Data Analysis

5.3.1 General Approach

Basic analyses of sediment data included:

- calculation of correlations within and among SQT components; and
- regression of sediment quality variables (*Y*) on distances from active drill centres.

Spearman's non-parametric rank correlation (r_s) was used for correlation analyses. Spearman's r_s is the parametric or Pearson correlation (r) between the ranks of two variables. Rank correlations are useful when there are values less than EQL and extreme values.

Distance (*X*) variables for the distance regressions were distances from the Northern, Central and Southern drill centres. These were considered "active" drill centres (see Section 4 for drill mud discharge statistics). Distances from the NN and SS drill centres were not considered because no drilling occurred there prior to completion of the sediment survey. Water column depth was also included as an *X* variable because the baseline survey showed that depth affected some variables (Husky Energy 2001)[b].

Directional effects were inferred from differences in the strength and sign of distance slopes among drill centres (a form of triangulation) and from bubble plots (spatial distributions, with the size of circles representing levels, or concentrations, of *Y* variables). Depth could also be considered a directional variable because depth increased to the northeast. More specific directional variables

[b] Depth was uncorrelated or weakly correlated with the three distance measures. Distances from the Northern and Southern drill centres were weakly negatively correlated. However, distances from the Central and Southern drill centres were strongly positively correlated because the two centres were close to each other. The regressions were based on partial sums-of-squares (SS), with effects of any *X* variable estimated after inclusion of (i.e., independent of the effects of) all other X variables. Using partial SS reduced or removed any confounding of the effects of distances from the Central and Southern drill centres. However, when two *X* variables are strongly correlated estimates of regression slopes may not be robust and may have wide confidence limits.

relative to each drill centre were not included because the objective was to simplify regressions by reducing the number of *X* variables.

Distances and depths (X variables) were $\log_{10}$ transformed for regressions. Except for multivariate summary measures, *Y* variables were also log-transformed.

Distance regressions were compared between 2000 (baseline) and 2004 (EEM). Appendix B-5 provides the Repeated Measures (RM) regression approach used, which compared depth and distance gradients between years for the 37 stations sampled in both years. Effectively, the RM regressions are regressions of the differences between years for each station versus depth and distances.

Statistical significance was defined based on the standard α, 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 $r_s| > 0.5$ $\{r^2 > 0.25\}$ and especially $|r$ or $r_s| > 0.7$ $\{r^2 \geq 0.5\}$); and
- large differences or changes over time or space (typically more than two-fold).

5.3.2 Physical and Chemical Characteristics

5.3.2.1 Groups of Variables

Four groups of related physical and chemical characteristics (*Y* variables) were examined:

- sediment particle size and Total Organic Carbon (TOC) content;
- major constituents of drilling muds and indicators of drilling activity (barium and $>C_{10}$-C_{21} HCs);
- frequently detected metals; and
- other inorganic compounds (sulphur, ammonia).

Except for the drilling indicators, the groups of *Y* variables analyzed can be considered to be:

- modifying or explanatory variables, potentially affecting other physical and chemical characteristics, toxicity test results, and benthic invertebrate communities; and
- potential low-level indicators that could be affected by drilling and other project activities.

Sediment particle size was expressed as % contributions of gravel, sand and fines (silt + clay). Both fines and TOC content could be elevated by drilling activity. Drilling muds are finer than the predominantly sand substrate on the Grand Banks.

Barium, as barium sulphate (barite), is a constituent of WBMs. Similarly, >C_{10}-C_{21} HCs are components of SBMs.

Other metals were treated largely as reference metals, or indicators of natural patterns that barium as a naturally-occurring metal would allow in the absence of drilling.

Sulphur, as sulphate in barite, is an important constituent of WBMs, although high background levels (parts per thousand) may obscure any increases from WBM use.

5.3.2.2 Statistical Analysis

Spearman rank correlations (r_s) were calculated within and among groups of variables. For the rank correlations, values less than EQL were treated as tied for the lowest rank.

Principal Components Analysis (PCA) was used to derive a summary measure of concentrations of nine metals (aluminum, chromium, iron, lead, manganese, strontium, uranium, vanadium and zinc) frequently detected in 2000 and 2004. PCA identifies the major axis of covariance (Principal Component 1 or PC1) among the original variables (i.e., concentrations of the nine metals), which is also the major axis of variance among stations. The minor axis (PC2) is the axis accounting for the largest amount of the remaining covariance or variance that is independent of (uncorrelated with) PC1. Positions of samples on the PC axes can be expressed as scores, and the scores used for subsequent analysis.

Metal concentrations were $\log_{10}$ transformed prior to conducting the PCA. All stations sampled in 2000 and 2004 were included, except for the two remote Reference stations sampled in 2000 (see Section 1). The PCA was conducted on the correlation, rather than the covariance, matrix. Of the nine metals summarized by PCA, zinc was the only metal to occur at concentrations less than EQL, and this only in 2004. Zinc concentrations below EQL were set at ½ EQL (or 2.5 mg/kg), which introduced an artificial source of variance. First, zinc concentrations at or above EQL varied little over space and time, so the two-fold difference between ½ EQL and EQL represented a relatively large difference on a log scale. Second, the EQL for zinc was 2 mg/kg in 2000 and 5 mg/kg in 2004, so there were some measurable concentrations less than 5 mg/kg in 2000. The effects of these artificial sources of variance were considered minimal because zinc was combined with eight other correlated metals in the robust summary measure, Metals PC1.

For the distance regressions for 2004, 11 >C_{10}-C_{21} HC concentrations less than EQL were set at ½ EQL (or 0.125 mg/kg). This could have introduced some artificial variance, but the two-fold difference between ½ EQL and EQL was trivial compared to the 1,000-fold differences among concentrations at or above EQL. There was also one sulphur concentration below an EQL of 0.02%. Setting that concentration at ½ EQL (or 0.01%) would have significantly inflated variances

since most concentrations were within a two-fold range between 0.02% (EQL) and 0.04%. Therefore, the one concentration below EQL was set at EQL.

Distance and depth regressions were strongly affected by six stations representing extreme values of *X* or *Y* variables. These stations were N4 and S5, located approximately 300 m from the Northern and Southern drill centres, and the four Reference stations (4, 12, 19 and 27). Therefore, regressions were calculated with and without these six stations.

5.3.3 Toxicity

Correlation and regression analyses were not conducted on toxicity test responses because no field sediments were toxic to amphipods or Microtox bacteria.

5.3.4 Benthic Community Structure

5.3.4.1 Groups of Variables

Benthic invertebrate 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
- relative abundances of major (higher-level) taxa.

Nemerteans, nematodes, oligochaetes, ostracods and copepods were excluded from all variables except standing crop because these small organisms are poorly recovered with the 0.5-mm mesh sieve used. These excluded organisms made a negligible contribution to standing crop because of their small size.

Major taxa analyzed were Polychaeta, Bivalvia, Amphipoda, Tanaidacea and Echinodermata; the five most abundant taxa. Relative abundances were major taxon abundances as a percent of total abundance.

5.3.4.2 Statistical Analysis

Preliminary Analysis

For 2000 (baseline) and 2004 (EEM) samples, abundances for each taxon for the two cores collected at each station were summed. Genera and species within families were pooled and families were used as the basic taxonomic unit for analysis of abundances, occurrence and other measures. The guidance manuals for the national pulp and paper, and mining EEM programs

(Environment Canada 2004; 2002b) provide practical rationales for pooling lower-level taxa to family or higher taxonomic levels. For the White Rose data, there was good agreement at the family level between the two taxonomists used in 2000 and 2004. At lower taxonomic levels, there were some differences in the level of taxonomic identifications (e.g., genus versus species) and in the treatment of uncertain identifications. Appendix B-5 provides abundances of lower-level taxa (usually species) in the 2004 samples, and summary measures based on those taxonomic levels.

Measures of richness, diversity, evenness and community composition were based on pooled abundances and occurrences of taxa at the family level. Richness (*S*) was the number of families per stations. Simpson's *D* was used as a diversity measure:

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

where p_i is the abundance of the *i*th taxon as a proportion of total abundance. *D* is the number of "very abundant" taxa (Ludwig and Reynolds 1988), with lower values indicating lower diversity. Simpson's evenness (*E*) is then *D*/*S*, or the number of very abundant taxa as a proportion of the total number of taxa. Although evenness is calculated from diversity, diversity is defined as consisting of two components: richness and evenness (i.e., *D*=*ExS*).

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 were expressed as a percent of total abundance. Second, Bray-Curtis (BC) similarities were calculated between all possible pairs of stations. These BC similarities are the percentage of invertebrates shared between stations (percent similarity). Third, the BC 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. Stress values less than or equal to 0.1 indicate good fits; stress values between 0.1 and 0.2 indicate adequate fits; stress values greater than 0.2 indicate poor fits (Clarke 1993). Positions of stations along the dimensions (MDS1, MDS2, etc.) or *scores* can then be used as summary measures of community composition for further analysis.

Correlation Analysis

For all 56 stations sampled in 2004, Spearman's rank correlations (r_s) were calculated:

- among the seven benthic invertebrate community summary measures: total abundance, standing crop, richness, diversity, evenness, MDS1 and MDS2 scores; and
- between summary measures and the relative abundances of major taxa.

Non-zero correlations were expected between many of these variables (e.g., between diversity and its two components, richness and evenness). In many cases, the primary objective was not to test expected correlations, but to indicate that results should be similar for correlated variables.

Distance and Depth Effects

The seven summary measures (*Y* variables) were regressed on water column depth and distances from the drill centres (*X* variables, see Section 5.3.1). Total abundance was $\log_{10}$ transformed. Other *Y* variables were not transformed. To be consistent with analysis of sediment physical and chemical characteristics, regressions were calculated for all 56 stations sampled in 2004, and then for a trimmed set of 50 stations (with N4, S5 and the four Reference stations excluded).

Rank correlations between major taxon abundances, depth and distances were also calculated for all 56 stations. Rank correlations remove the effects of extreme *Y* or *X* values, so analysis of the trimmed data set was unnecessary in this case.

The RM regression model described in Appendix B-5 was used to compare the seven benthic invertebrate community summary measures between 2000 and 2004. Rank correlations between relative abundances of major taxa, depth and distance were also compared between years.

5.3.5 Integrated Assessment

Integrated assessment of SQT components consisted of calculating bivariate correlations (r_s) between selected physical characteristics, chemical characteristics and benthic invertebrate community variables.

5.4 Results

In the description of results that follows, reference to positive and negative correlations with distance from drill centres indicates increases (positive correlation) of a given variable with increasing distance from the centres, or decreases (negative correlation) of variables with increasing distance from centres. Positive or negative correlations among groups of variables are also discussed. Again, a positive correlation indicates increasing levels of one variable with increasing levels of another; while a negative correlation indicates decreasing levels of one variable with increasing levels of another.

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 and 2004. Table 5-3 (Section 5.2) provides a list of all chemical characteristics measured in 2004. BTEX was not detected in sediment in both 2000 and 2004. $>C_{10}$-C_{21} and $>C_{21}$-C_{32} HCs were not detected in sediments in 2000 but were detected in

2004. One PAH, naphthalene, was detected in one sample in 2000. Of the metals, arsenic was detected in 13 samples in 2000 but was not detected in 2004. Antimony, beryllium, mercury, molybdenum, selenium and tin were not detected in sediments in either year.

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

Variable	Year	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV
$>C_{10}$-C_{21}	2000	46	46	<0.25	<0.25	<0.25			
	2004	56	11	<0.25	275.00	0.74			
$>C_{21}$-C_{32}	2000	46	46	<0.25	<0.25	<0.25			
	2004	56	45	<0.25	0.92	<0.25			
(C_6-C_{32})	2000	46	46	<0.25	<0.25	<0.25			
	2004	56	44	<3	276.00	<3			
Naphthalene	2000	46	45	<0.05	0.07	<0.05			
	2004	56	56	<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
TOC	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
TIC	2000	46	6	<0.1	0.40	0.10			
	2004	56	52	<0.3	0.50	<0.3			
Aluminum	2000	46	0	6400	11000	8250	8243	651	8
	2004	56	0	6500	9500	8300	8173	709	9
Arsenic	2000	46	33	<2	2.00	<2			
	2004	56	56	<2	<2	<2			
Barium	2000	46	0	120.00	210.00	160.00	163.70	19.36	12
	2004	56	0	110.00	1400.00	160.00	203.39	177.66	87
Cadmium	2000	46	46	<0.05	<0.05	<0.05			
	2004	56	38	<0.05	0.08	<0.05			
Chromium	2000	46	0	3.00	4.00	3.00	3.46	0.50	15
	2004	56	0	3.00	7.00	4.00	3.80	0.70	18
Cobalt	2000	46	44	<1	1.00	<1			
	2004	56	50	<1	1.00	<1			
Copper	2000	46	41	<2	4.00	<2			
	2004	56	19	<2	3.00	<2			
Iron	2000	46	0	1100	2300	1400	1461	244	17
	2004	56	0	850	2400	1500	1489	315	21
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
Lithium	2000	46	46	<5	<5	<5			
	2004	56	31	<2	2.00	<2			
Manganese	2000	46	0	25.00	70.00	36.00	38.65	10.12	26
	2004	56	0	17.00	82.00	38.00	40.05	12.65	32
Nickel	2000	46	44	<2	2.00	<2			
	2004	56	54	<2	2.00	<2			
Strontium	2000	46	0	37.00	60.00	47.00	47.48	3.49	7
	2004	56	0	34.00	64.00	46.00	47.00	4.87	10

Variable	Year	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV
Thallium	2000	46	1	<0.1	0.10	0.10			
	2004	56	0	0.10	0.10	0.10	0.10	0.00	0
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
Vanadium	2000	46	0	5.00	8.00	6.00	6.41	0.69	11
	2004	56	0	4.00	7.00	6.00	5.71	0.76	13
Zinc	2000	46	0	4.00	14.00	6.00	6.41	2.27	35
	2004	56	10	<5	9.00	<5			
Ammonia	2000	NA							
	2004	56	0	2.17	64.60	7.10	9.23	9.00	98
%(w) Sulphur	2000	NA							
	2004	56	1	<0.02	0.08	0.03			
Sulphide	2000	NA							
	2004	56	53	<2	3.00	<2			
% 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
% 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
% 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
% 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
% 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

Notes:
- 2000 data exclude two remote Reference stations (see Section 1)
- Metal, ammonia, sulphur and sulphide concentrations are in mg/kg dry wt
- Total carbon, TOC and TIC are in g/kg
- NA = Not analyzed

5.4.1.1 Correlations Within and Among Groups of Variables (2004)

Correlations among sediment particle size categories and TOC content for the 56 stations sampled in 2004 are provided in Table 5-5. The sediments were predominantly sand, with low gravel, fines and TOC content. 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 categories sum to 100%. Gravel content, which was usually the major non-sand component, was strongly negatively correlated with sand content. Fines content, the minor non-sand component, was weakly negatively correlated with sand content. Gravel and fines content were uncorrelated. These results indicate that finer particles occurred or were deposited in both sand and gravel, rather than primarily in the interstitial spaces in gravel. TOC content was also uncorrelated with fines content and weakly positively correlated with gravel content. Therefore, organic carbon did not appear to be associated with finer particles.

Table 5-5 Spearman Rank Correlations (r_s) Among Particle Size and Organic Carbon Content (2004)

	% fines	% sand	% gravel
% sand	–0.378**		
% gravel	0.085	**–0.934*** **	
TOC	0.150	–0.321*	0.273*

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

Concentrations of the two primary drilling mud indicators, barium (in WBMs) and >C_{10}-C_{21} HCs (in SBMs), were positively correlated (Table 5-6). A positive correlation was expected since both types of drilling muds were used at the Northern and Southern drill centres and concentrations of both indicators would be expected to be low at more remote stations.

Table 5-6 Spearman Rank Correlations (r_s) Among Barium and HC Concentrations (2004)

	Barium	>C_{10}-C_{21} HCs
>C_{10}-C_{21} HCs	**0.464*** **	
>C_{21}-C_{32} HCs	0.073	0.352**

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

Table 5-6 also includes correlations with >C_{21}-C_{32} HCs, which were detected at 11 stations. Concentrations of >C_{21}-C_{32} and >C_{10}-C_{21} HCs were positively correlated, indicating that the few >C_{21}-C_{32} HC concentrations above EQL generally occurred at stations where >C_{10}-C_{21} HC concentrations were high.

Concentrations of the nine frequently detected metals in sediments collected in 2000 and 2004 were positively correlated with each other and with the first Principal Component (Metals PC1) derived from those concentrations (Table 5-7). PC1 accounted for more than 50% of the total variance and scores were used as a summary measure of metals concentrations. PC2 was negatively correlated with lead and zinc concentrations, but accounted for a limited amount of variance and was not retained for further analysis.

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

Metal	Correlation (*r*) with:	
	PC1	PC2
Iron	**0.896**	0.291
Strontium	**0.881**	–0.186
Aluminum	**0.874**	–0.160
Manganese	**0.823**	0.367
Vanadium	**0.681**	0.066
Lead	**0.671**	**–0.527**
Chromium	**0.636**	0.239
Zinc	**0.580**	**–0.525**
Uranium	0.446	0.453
% variance	54.2	12.2

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 = 102 stations; 56 in 2004 and 46 in 2000

Table 5-8 provides correlations between Metals PC1 scores and concentrations of three metals (cadmium, copper and lithium) detected at more than 10 stations in 2004. Those correlations increased in strength as the number of concentrations greater than EQL increased (i.e., as the data became more quantitative and robust), suggesting that most metals followed similar spatial patterns.

Table 5-8 Spearman Rank Correlations (r_s) Between Infrequently Detected Metals and Metals PC1 (2004)

Metal	Correlation (r_s) with Metals PC1
Cadmium (18 concentrations ≥ EQL)	–0.170
Copper (37 concentrations ≥ EQL)	**0.584*** **
Lithium (25 concentrations ≥ EQL)	0.343*

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

Ammonia concentrations were uncorrelated with drilling indicators, metal and sulphur concentrations (Table 5-9). As noted above, barium and $>C_{10}$-C_{21} HC concentrations were positively correlated. Sulphur concentrations were also positively correlated with barium and $>C_{10}$-C_{21} HC concentrations (Table 5-9), suggesting that sulphur (from barium sulphate in WBMs) might also be a drilling mud indicator. Barium and sulphur, but not $>C_{10}$-C_{21} HC, concentrations were also positively correlated with Metals PC1. In the absence of drilling effects, concentrations of barium and elements such as sulphur that occur naturally would be expected to co-vary with concentrations of other metals (i.e., barium and sulphur should "behave" like other metals and elements).

Table 5-9 Spearman Rank Correlations (r_s) Among Chemistry Variables

	Barium	>C_{10}-C_{21} HCs	Metals PC1	Sulphur
>C_{10}-C_{21} HCs	**0.464*** **			
Metals PC1	**0.687*** **	0.108		
Sulphur	**0.554*** **	0.407**	0.365**	
Ammonia	0.069	–0.010	0.126	–0.009

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

Except for ammonia, the chemistry variables were weakly positively correlated with sediment fines and TOC content (Table 5-10). Stronger correlations between metals, HCs and finer organic particles would normally be expected. However, and as noted elsewhere, there was limited variance of fines and TOC content, and levels of both were low.

Table 5-10 Spearman Rank Correlations (r_s) Between Chemistry Variables, Fines and TOC Content

Chemistry variable	Correlation (r_s) with:	
	% fines	TOC
>C_{10}-C_{21} HCs	0.118	0.148
Barium	0.393**	0.217
Metals PC1	0.279*	0.189
Sulphur	0.145	0.310*
Ammonia	–0.048	–0.107

Note: - *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$

5.4.1.2 Depth and Distance Effects (2004)

Extreme and Excluded Stations

Table 5-11 provides variable values for stations representing extremes of *X* (depth, distance) and *Y* variable values. The stations listed include the four Reference stations, the three stations located within approximately 300 m of the Northern, Southern and Central drill centres, plus the stations nearest to these 300-m stations. Table 5-11 also provides minimum, maximum and median *Y* and *X* values for all 56 stations, and for the trimmed data set with the four Reference stations and stations N4 and S5 excluded. Those six stations accounted for 9 of 14 minima and maxima for *Y* variables, and all minima and maxima for depth and distance from the nearest drill centre.

Table 5-11 Physical and Chemical Variable Values for All Stations, the Trimmed Set of Stations and Extreme (Near and Far) Stations

Data set	Statistic/ Station	Distance[1] (km)	Depth (m)	*Y* variable						
				Fines (%)	TOC (g/kg)	$>C_{10}$-C_{21} HCs (mg/kg)	Barium (mg/kg)	Sulphur (%)	Metals PC1	Ammonia (mg/kg)
All stations (*n* = 56)	Minimum	0.30	108	1.18	0.6	<0.25	110	<0.020	–2.59	2.17
	Maximum	26.19	175	3.26	1.2	275	1,400	0.082	2.50	64.6
	Median	3.08	127	1.44	0.95	0.74	160	0.027	–0.01	7.10
Trimmed (*n* = 50)	Minimum	0.33	119	1.20	0.8	**<0.25**	120	0.020	**–2.59**	**2.17**
	Maximum	16.00	139	2.10	**1.2**	37	470	0.043	1.89	**64.6**
	Median	2.97	127	1.43	1.0	0.79	160	0.027	–0.01	7.50
Near Northern	*N4*	**0.30**	126	1.67	1.0	8.99	240	0.036	0.24	4.94
	N3	0.63	126	1.30	0.9	27.7	190	0.031	–1.82	7.50
Near Southern	*S5*	0.31	127	**3.26**	0.9	**275**	**1,400**	**0.082**	**2.50**	6.57
	13	0.59	127	1.54	0.9	19.6	440	0.031	1.86	8.56
	S1	0.60	127	1.80	1.0	37	390	0.035	0.79	4.40
Near Central	C5	0.33	124	1.30	1.0	0.49	160	0.035	0.28	5.30
	20	0.35	127	1.20	0.9	1.05	140	0.029	–0.97	4.94
	C3	0.74	123	1.30	0.9	0.78	150	0.029	–0.13	6.34
References	*4 (NE)*	**26.19**	**175**	2.91	0.9	**<0.25**	180	0.021	1.30	6.75
	12 (SE)	25.85	137	1.70	0.9	0.43	140	0.027	–0.34	6.89
	19 (SW)	26.18	**108**	**1.18**	**0.6**	**<0.25**	**110**	0.023	–2.09	9.16
	27 (NW)	20.03	123	1.28	0.8	0.35	140	**<0.020**	–2.13	6.40

Notes:
- Italics indicate stations that were trimmed (N4, S5 and Reference stations)
- Bold indicates minima or maxima for all 56 stations that occurred either in the trimmed data set or at the extreme stations
- [1] Distance to nearest active drill centre (Northern, Southern, Central)

Station S5 was usually a positive outlier, with high values for most *Y* variables (Table 5-11). *Y* values also tended to be high at station N4. Both S5 and N4 represented extreme values of distance from the Northern or Southern drill centres, or the nearest of the two centres, on a log scale. For these two stations, the important issue is whether any apparent effects there extended beyond 300 m from the drill centres. The Reference stations were excluded because stations 4 and 19 represented depth extremes, and regressions on depth were largely two-point regressions between those two values. For consistency, all four Reference stations were excluded, even though stations 12 and 27 did not represent depth extremes. On a log scale, none of the Reference stations represented distance extremes.

Regression Analysis

Tables 5-12 and 5-13 provide results of regressions on depth and distance for the full data set of 56 stations and the trimmed data set of 50 stations. When results differ between the two data sets, one can infer that the excluded stations in Table 5-11 were largely responsible for any apparent effects observed for the full data set. When *Y* variables occurred at elevated levels at stations N4 and S5, but also occurred at elevated levels at the next nearest stations in Table 5-11, then the regressions for the trimmed data set were usually significant (Table 5-13).

Table 5-12 Results of Regressions of Physical and Chemical Variables on Depth and Distance from Active Drill Centres for All Stations (2004)

Model/term	Result	*Y* variable						
		% fines	TOC	$>C_{10}$-C_{21} HCs	Barium	Sulphur	Metals PC1	Ammonia
X = Depth + distances (*d*) from all drill centres								
Overall	r^2	**0.515*** **	0.230**	**0.688*** **	**0.700*** **	**0.357*** **	**0.358*** **	0.020
Depth	Slope *(b)*	**2.134*** **	0.662*	–2.157	1.053	0.187	17.232**	–0.619
Northern *d*	Slope *(b)*	–0.028	–0.035*	**–0.644*** **	–0.102**	–0.077**	–0.264	0.024
Southern *d*	Slope *(b)*	**–0.117*** **	–0.049*	**–1.869*** **	**–0.439*** **	**–0.152*** **	**–1.520*** **	–0.077
Central *d*	Slope *(b)*	0.084**	0.035	**0.965*** **	**0.211*** **	0.057	0.214	0.040
X = Depth + distances (*d*) from Northern and Southern drill centres								
Overall	r^2	**0.427*** **	0.181*	**0.527*** **	**0.554*** **	**0.319*** **	**0.355*** **	0.017
Depth	Slope *(b)*	**2.368*** **	0.759**	0.534	1.642*	0.345	**17.829*** **	–0.507
Northern *d*	Slope *(b)*	–0.011	–0.028	–0.449*	–0.059	–0.066*	–0.221	0.032
Southern *d*	Slope *(b)*	–0.064**	–0.027	**–1.259*** **	**–0.306*** **	**–0.116*** **	**–1.384*** **	–0.051
X = Distance from nearest drill centre								
Overall	r^2	0.001	0.012	**0.350*** **	**0.262*** **	**0.284*** **	0.089*	0.003
Distance	Slope *(b)*	–0.006	–0.012	**–0.934*** **	**–0.185*** **	**–0.103*** **	–0.686*	0.025
X = Distance from nearest of Northern and Southern drill centres								
Overall	r^2	0.038	0.066	**0.593*** **	**0.456*** **	**0.361*** **	0.139**	0.000
Distance	Slope *(b)*	–0.041	–0.030	**–1.379*** **	**–0.277*** **	**–0.131*** **	–0.972**	–0.001

Notes:
- *$p \le 0.05$; **$p \le 0.01$; ***$p \le 0.001$ (in bold)
- For regressions with a single *X* (distance) variable, *p* are the same for r^2 and slopes (*b*)
- All *X* variables and all *Y* variables except Metals PC1 were $\log_{10}$ transformed

Table 5-13 Results of Regression of Physical and Chemical Variables on Depth and Distances from Active Drill Centres for the Trimmed Data Set of 50 Stations (2004)

Model/term	Result	*Y* variable						
		% fines	TOC	$>C_{10}$-C_{21} HCs	Barium	Sulphur	Metals PC1	Ammonia
X = Depth + distances (*d*) from all drill centres								
Overall	r^2	**0.364*** **	0.224*	**0.665*** **	**0.708*** **	0.173	0.186	0.027
Depth	Slope *(b)*	**2.621*** **	0.686	–10.029	0.980	1.088	16.132	–1.486
Northern *d*	Slope *(b)*	–0.026	0.003	**–0.808*** **	–0.111**	–0.052	–0.085	–0.047
Southern *d*	Slope *(b)*	–0.078*	–0.030	**–1.972*** **	**–0.379*** **	–0.076*	–1.272*	–0.136
Central *d*	Slope *(b)*	0.045	0.048*	**0.943*** **	**0.146*** **	0.003	0.085	0.079
X = Depth + distances (*d*) from Northern and Southern drill centres								
Overall	r^2	**0.324*** **	0.115	**0.492*** **	**0.588*** **	0.173*	0.185*	0.018
Depth	Slope *(b)*	**3.065*** **	1.154*	–0.787	2.408*	1.119	16.962	–0.711
Northern *d*	Slope *(b)*	–0.015	0.015	–0.580*	–0.076	–0.051	–0.065	–0.028
Southern *d*	Slope *(b)*	–0.050*	–0.001	**–1.405*** **	**–0.292*** **	–0.074**	–1.221**	–0.089
X = Distance from nearest of Northern and Southern drill centres								
Overall	r^2	0.026	0.002	**0.571*** **	**0.479*** **	0.169**	0.076	0.003
Distance	Slope *(b)*	–0.035	–0.007	**–1.654*** **	**–0.281*** **	–0.084**	–0.896	–0.043
X = Distance from Southern drill centre								
Overall	r^2	0.009	0.002	**0.436*** **	**0.505*** **	0.076	0.143**	0.016
Distance	Slope *(b)*	–0.017	0.005	**–1.229*** **	**–0.245*** **	–0.048	–1.043**	–0.086

Notes:
- Trimmed data set excludes four References stations (4, 12, 19, 27) and N4 and S5 (see Table 5-11 for values or excluded stations)
- *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$ (in bold)
- For regressions with a single *X* (distance) variable, *p* are the same for r^2 and slopes (*b*)
- All *X* variables and all *Y* variables except Metals PC1 were $\log_{10}$ transformed

Tables 5-12 and 5-13 provide two measures of effect size: r^2 and slopes (*b*). The r^2 provide the proportion of the total variance of *Y* accounted for by the models. Regression slopes (*b*) measure the rate of change in *Y* with *X*. Steeper negative distance slopes indicate greater attenuation with distance, which can be an indicator of greater effects from drilling near drill centres.

Slopes for distance from the Central drill centre were positive and often significant, especially for the set of all 56 stations (Tables 5-12 and 5-13). Distance from the Central drill centre was primarily a localized directional variable, with positive slopes indicating more rapid decreases in *Y* values near the Southern drill centre towards the Central drill centre (northwest) than to the southeast or in other directions (see below). Despite the strong correlation between distances from the Central and Southern drill centre, and the possible risk of confounding effects of the two distances, the regression model and results for distance from the Central drill centre identified directional effects independent of distance from the Southern drill centre that were visually evident in bubble plots.

Because slopes for distances from the Northern and Southern drill centres were usually negative, and slopes for distance from the Central drill centre were usually positive, distance from the nearest of the three drill centres was never as effective as a single *X* variable as distance from the nearest of just the Northern and Southern drill centres (compare r^2 in Table 5-12).

For all stations, there were significant increases in fines content with depth (Table 5-12 and Figure 5-6). Those effects were significant even when the two extreme depths at stations 4 and 19 were excluded (Table 5-13 and Figure 5-6). The highest fines content occurred at station S5 near the Southern drill centre (Table 5-11). However, slopes for distance from that drill centre were significant in models excluding S5 but including depth effects (Table 5-13), partly because the high fines content at Reference station 4 were also excluded. Distance from the Northern drill centre had no apparent effects (Tables 5-12 and 5-13). Overall, fines content was higher near the Southern drill centre, especially at station S5 and otherwise increased to the Northeast with increasing depth (Figure 5-7). Fines content was also higher around the NN drill centre than at most other stations.

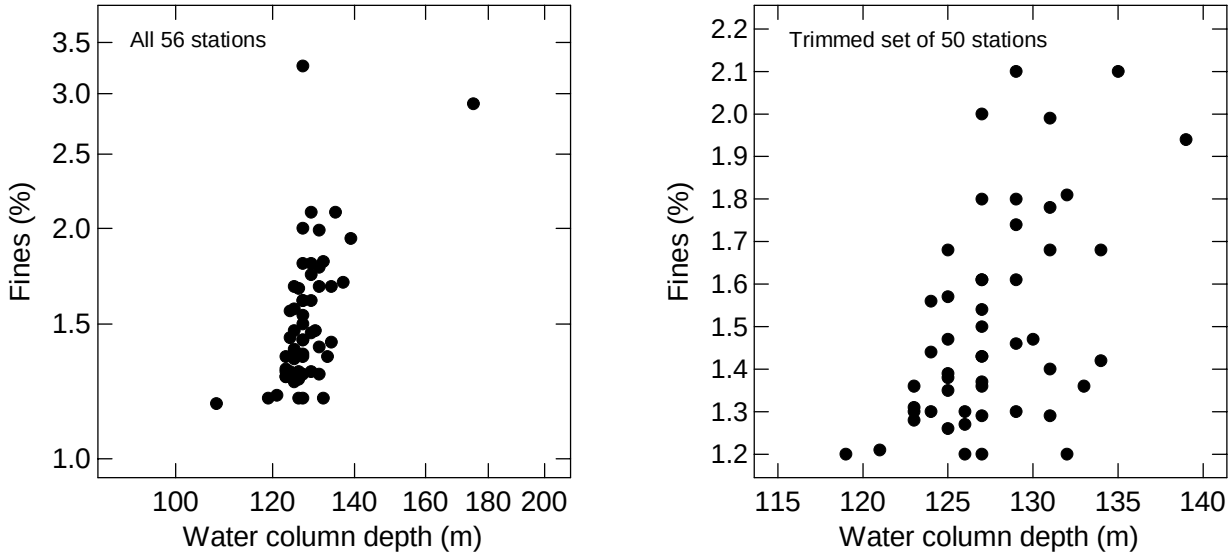


Figure 5-6 Sediment Fines Content Versus Depth (2004)

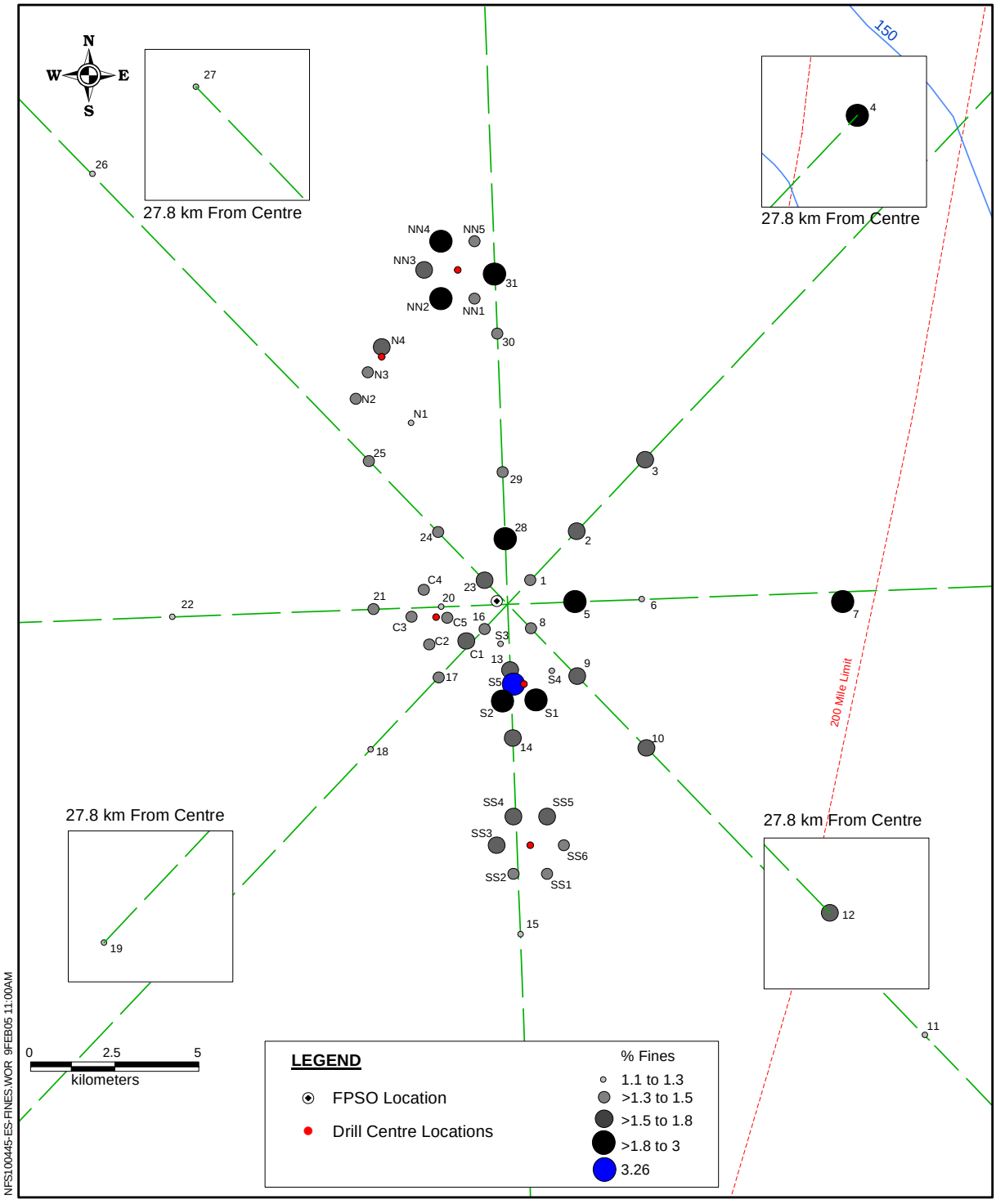


Figure 5-7 Spatial Distribution of % Fines (2004)

Sediment TOC content may have increased with depth and decreased with distance from the Northern and Southern drill centres (Tables 5-11 to 5-13), but any effects of depth and distance were weak given the limited range of TOC values (usually 0.9 or 1.0 g/kg). Overall, TOC content was marginally higher around the Southern, NN and SS drill centres and in the centre of the development area, and lowest (0.6 g/kg) at station 19 (SW Reference station) (Figure 5-8).

Concentrations of $>C_{10}$-C_{21} HCs decreased significantly with distance from the Northern and Southern drill centres, regardless of the distance variables or data set analyzed (Tables 5-12 and 5-13). In 2004, the median concentration of $>C_{10}$-C_{21} HCs was 22 mg/kg (range 9 to 275 mg/kg) within 1 km of the Northern and Southern drill centres, and levels fell to approximately 1 mg/kg at distances of 5 km from these drill centres. (Figure 5-9). Chromatograms for approximately 75% of the stations sampled within 8 km of the Northern or Southern drill centres had UCMs in the PureDrill IA-35 range (see Section 5.2.1, Figure 5.4 and Appendix B-3). Low levels of $>C_{10}$-C_{21} HCs were detected a three stations located more than 8 km from the drill centres (stations 11, 12, and 27; HC range: 0.42 and 0.66 mg/kg). However, these HCs did not have UCMs in the range of Puredrill IA-35 and PSC Maxxam reports that these HCs are probably non-petrogenic material.

Concentrations of $>C_{10}$-C_{21} HCs were higher around the Southern drill centre than around the Northern drill centre (Table 5-11; Figure 5-10), and slopes (attenuation with distance) were steeper for the Southern drill centre (Tables 5-12 and 5-13). Slopes for distance from the Central drill centre were significantly positive even for the trimmed data set (Table 5-13). $>C_{10}$-C_{21} HC concentrations near the Southern drill centre decreased more rapidly with distance towards the Central drill centre (northwest) than to the southeast (Figure 5-10).

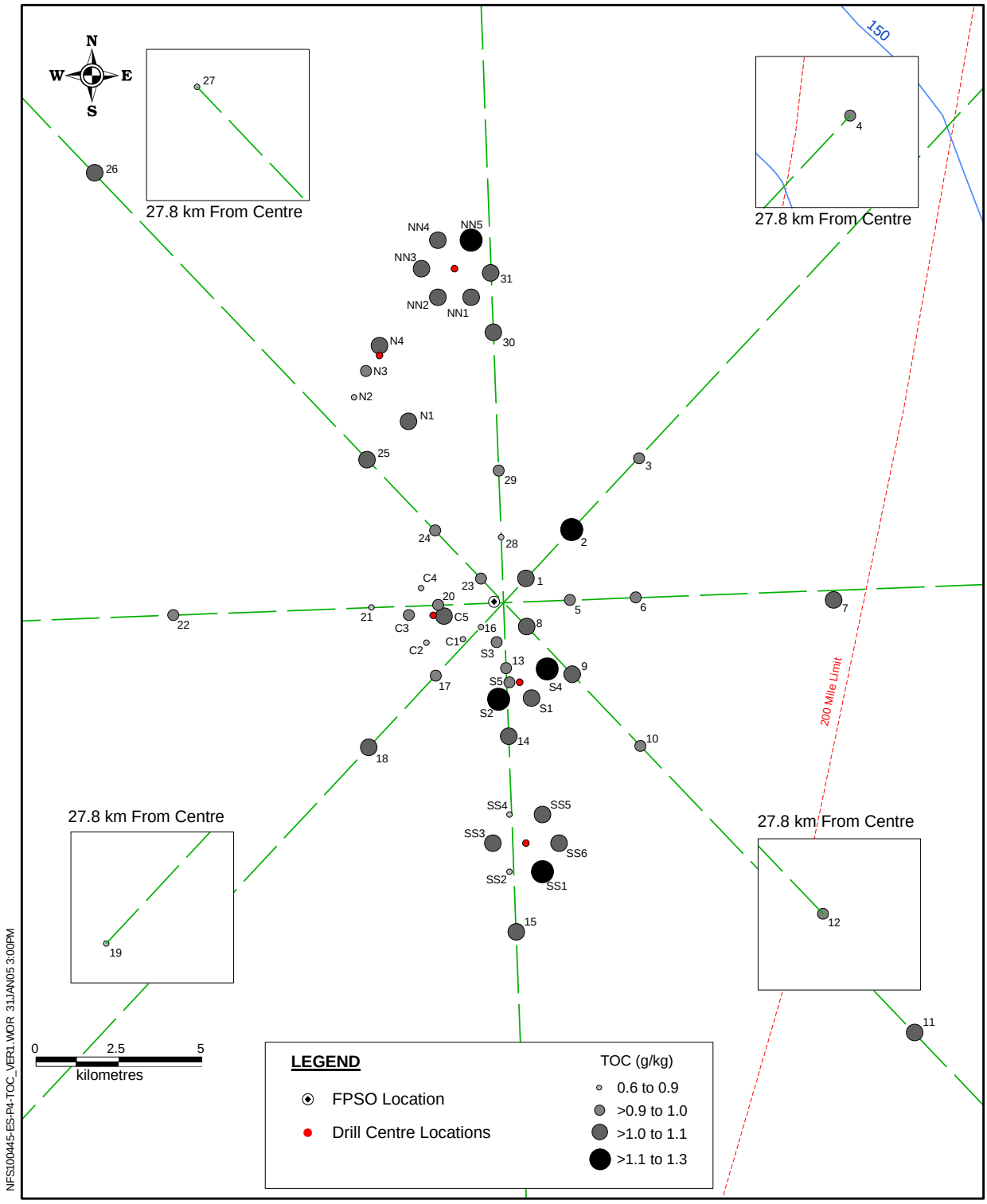


Figure 5-8 Spatial Distribution of TOC (2004)

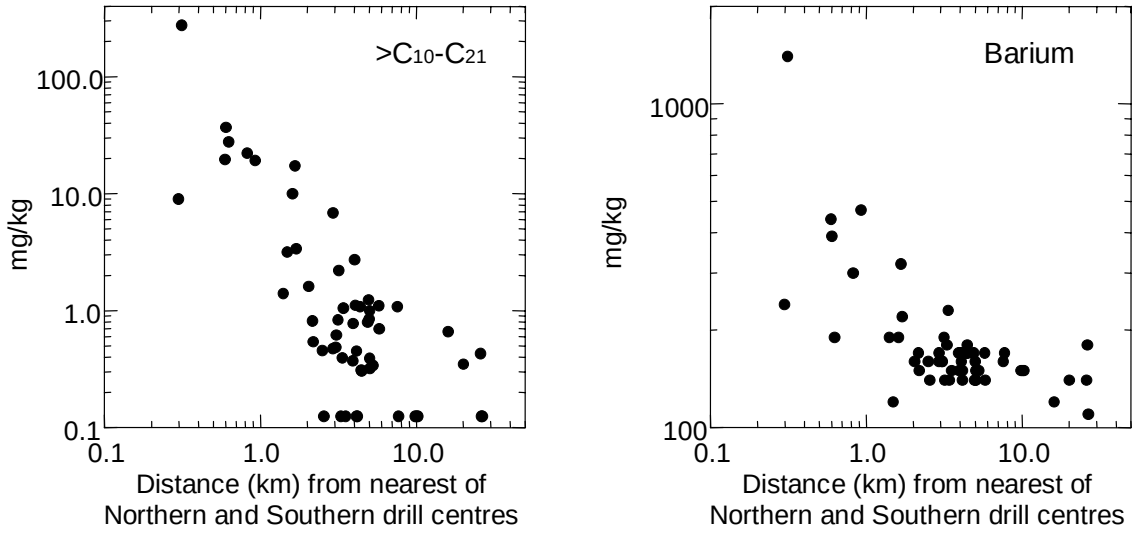


Figure 5-9 $>C_{10}$-C_{21} HCs and Barium Versus Distance from Drill Centres (2004)

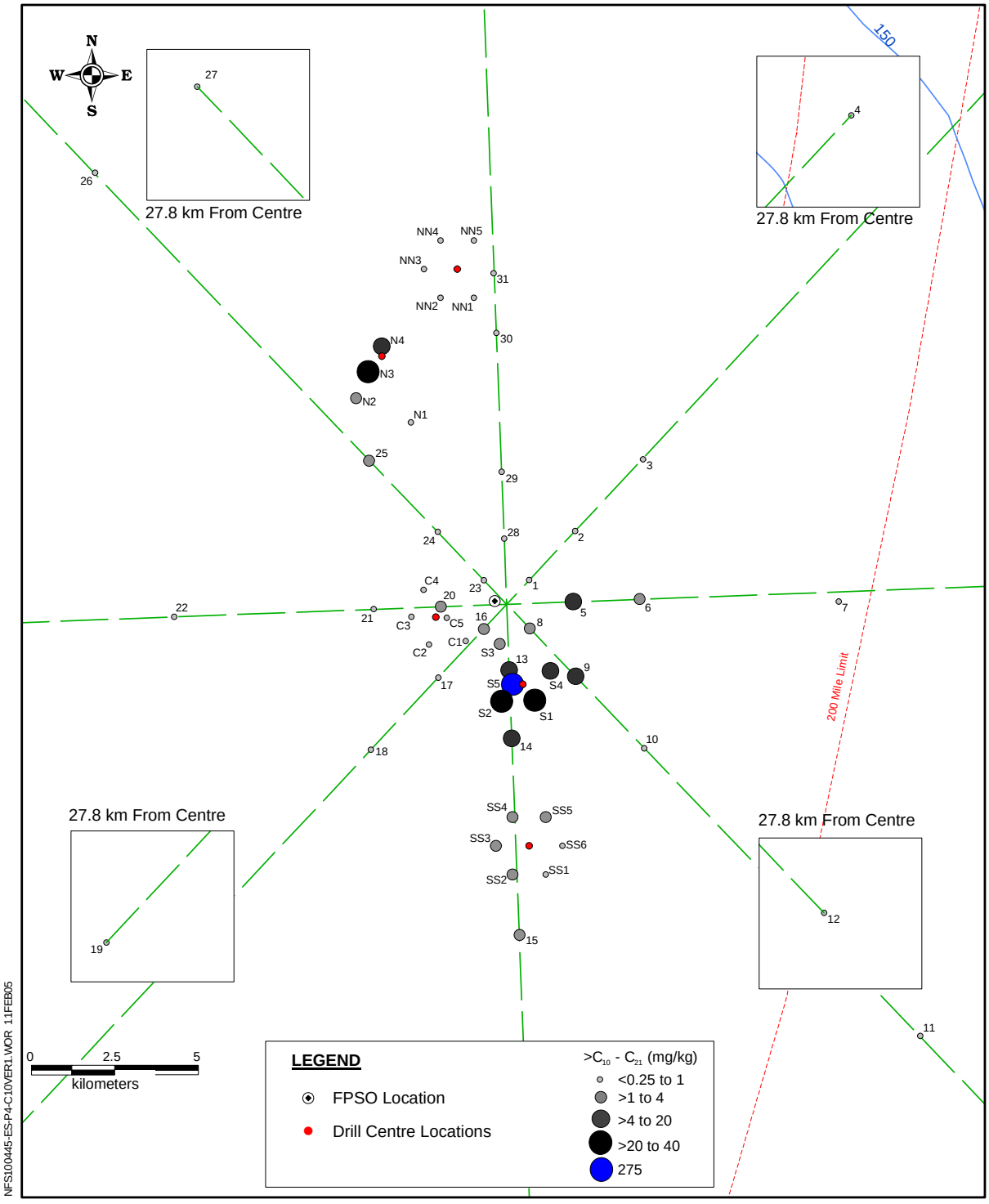


Figure 5-10 Spatial Distribution of >C_{10}-C_{21} HCs (2004)

Results for barium were similar to those for >C_{10}-C_{21} HCs, with concentrations decreasing with distance from the Northern and, especially, the Southern drill centres (Tables 5-11 to 5-13; Figure 5-9). The effects of the Northern drill centre are significant in the regression models primarily because concentrations near there were higher than expected based on medians or distance from the Southern drill centre. Most of the decreases in barium concentrations occurred within 2 km of either the Southern or Northern drill centres (also see Appendix B-5). The highest concentrations occurred at station S5 and nearby stations (Figure 5-11). Barium concentrations near the Southern drill centre also decreased more rapidly towards the Central drill centre (northwest) than to the southeast.

Sulphur concentrations at station S5 were approximately double concentrations at other stations (Table 5-11; Figure 5-12). At those other stations, concentrations decreased slightly with distance from the Northern and Southern drill centres (Table 5-13; Figure 5-13). Sulphur concentrations near the Southern drill centre did not decrease more rapidly towards the Central drill centre than in other directions and the effects of distance from the Central drill centre were not significant (Tables 5-12 and 5-13).

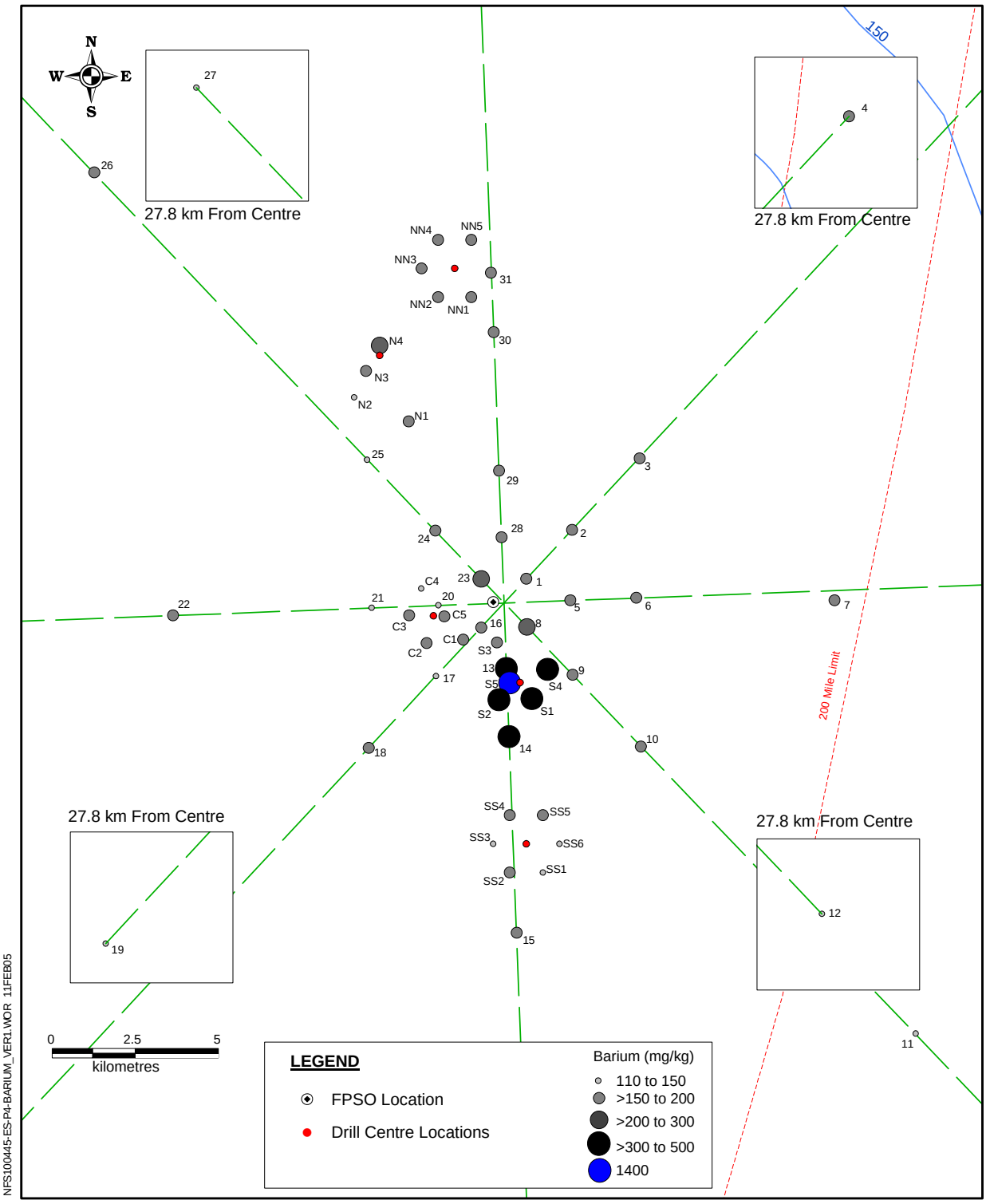


Figure 5-11 Spatial Distribution of Barium (2004)

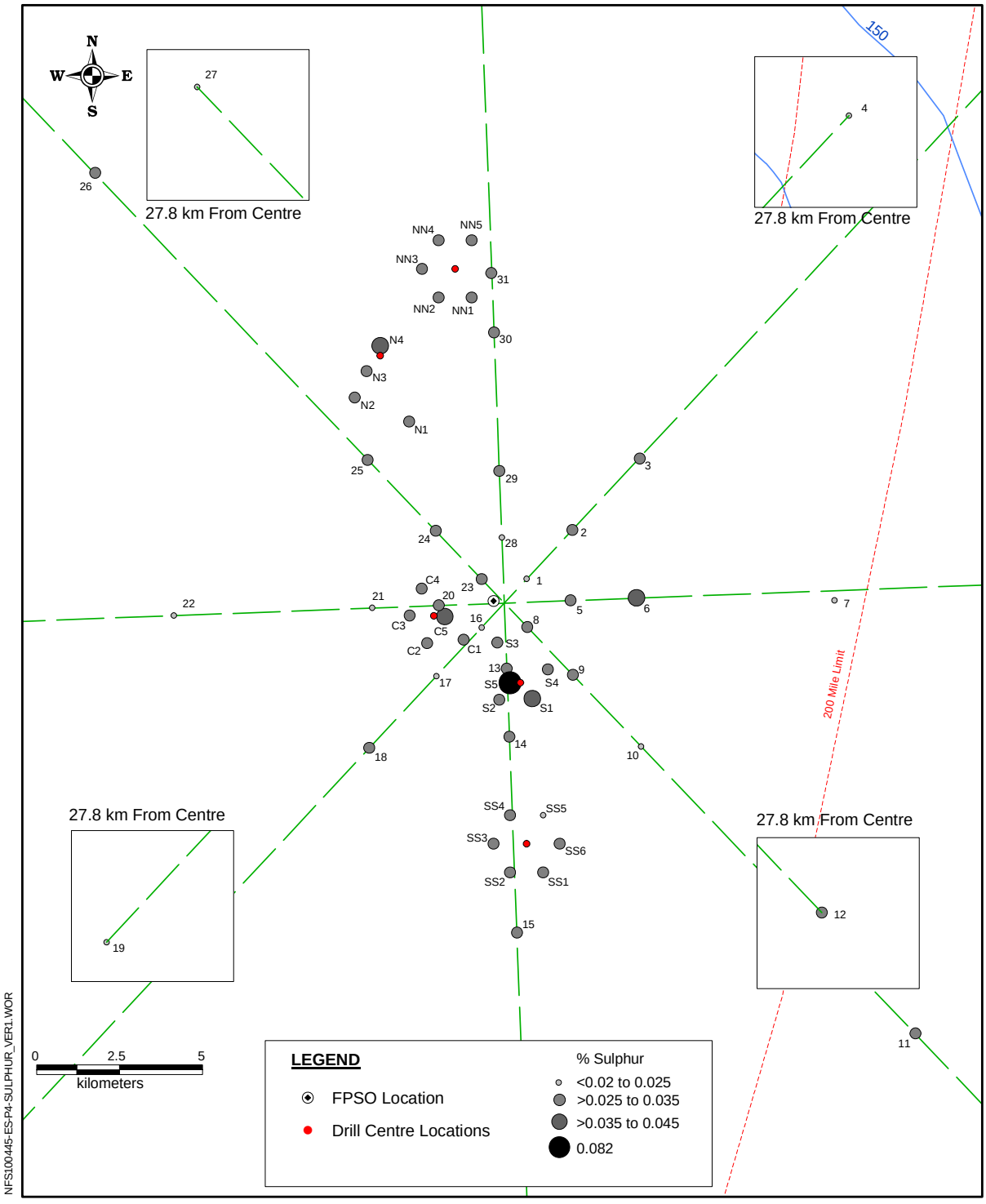


Figure 5-12 Spatial Distribution of Sulphur (2004)

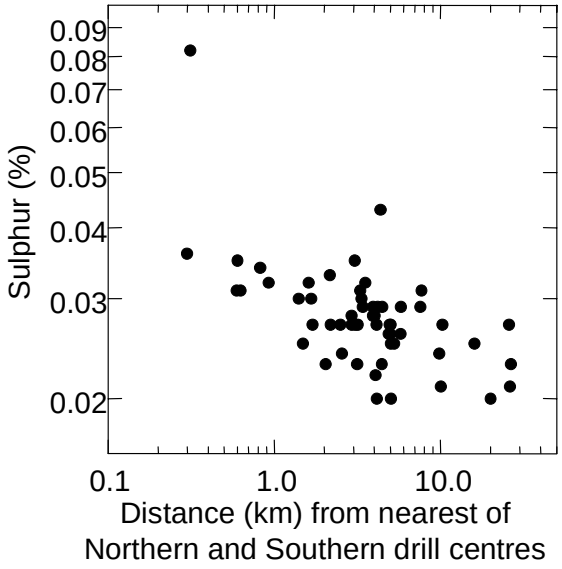


Figure 5-13 Sulphur Versus Distance from Drill Centers (2004)

Metal concentrations and Metals PC1 scores were highest at station S5 and surrounding stations (Table 5-11; Figure 5-14) and decreased significantly with distance from the Southern drill centre (Tables 5-12 and 5-13; Figure 5-15). Concentrations also decreased with distance from the Northern drill centre. However, those decreases were not significant for either the full or trimmed data sets (Tables 5-12 and 5-13). Distance from the Southern drill centre alone was a better predictor of metal concentrations than distance from the nearest of the Northern and Southern drill centres (Table 5-13). Metal concentrations also increased with depth, but that increase was significant only when stations 4 and 19 (extreme depth values) were included in regressions (Tables 5-12 and 5-13).

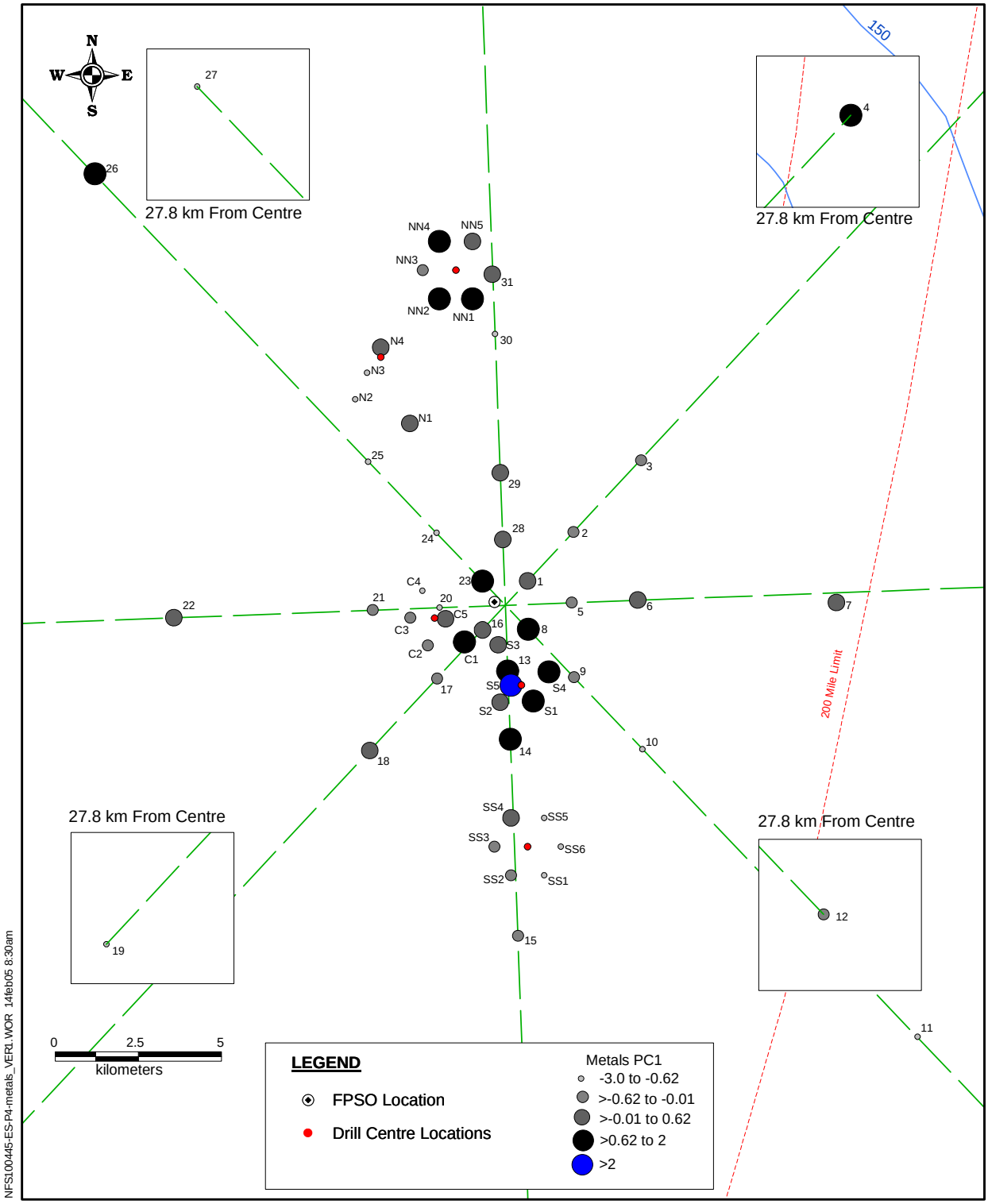


Figure 5-14 Spatial Distribution of Metals PC1 (2004)

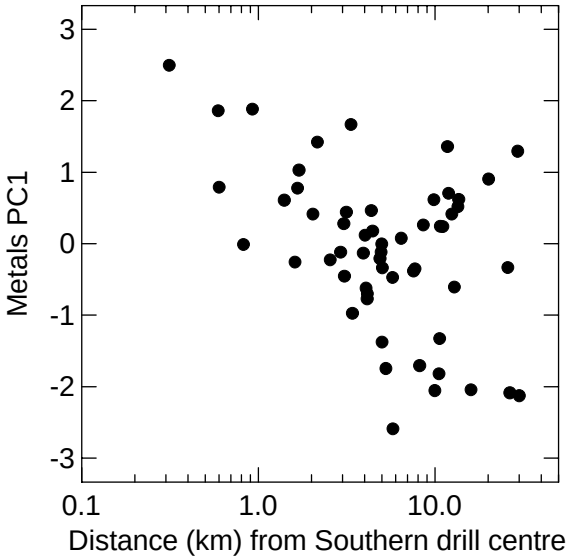


Figure 5-15 Metals PC1 Scores Versus Distance from the Southern Drill Centre (2004)

Ammonia concentrations were unrelated to depth and distance from the drill centres (Tables 5-12 and 5-13). The highest concentration (64.6 mg/kg) occurred at station SS6, near the proposed SS drill centre (Figure 5-16).

5.4.1.3 Comparison Between Years (2000 and 2004)

Results of RM regression analyses of sediment physical and chemical characteristics for the 37 stations sampled in both 2000 and 2004 are provided in Table 5-14. Appendix B-5 provides details on interpretations of the terms in the models. The Among Stations terms test for effects common to both years (i.e., effects on mean *Y* over the two years). The Within Stations Terms (Between Years) test differences between years. The Year term tests for differences between years common to most stations. The Year $\times$ *X* terms test for differences in depth or distance effects (i.e., slopes) between years. Results are provided as *F* values, which are a measure of effect size. *F* will be greater than 1 when there is added variance attributable to the effect or term tested. Among Stations effects of *X* variables common to both years (effects on means) should usually be ignored when Within Stations effects for the same variable (effects on differences, or differences in effects) are of similar magnitude (*F*) or significance. When effects differ between years, testing or calculating common or average effects over both years is of questionable value (i.e., differences are more relevant than the averages of those differences).

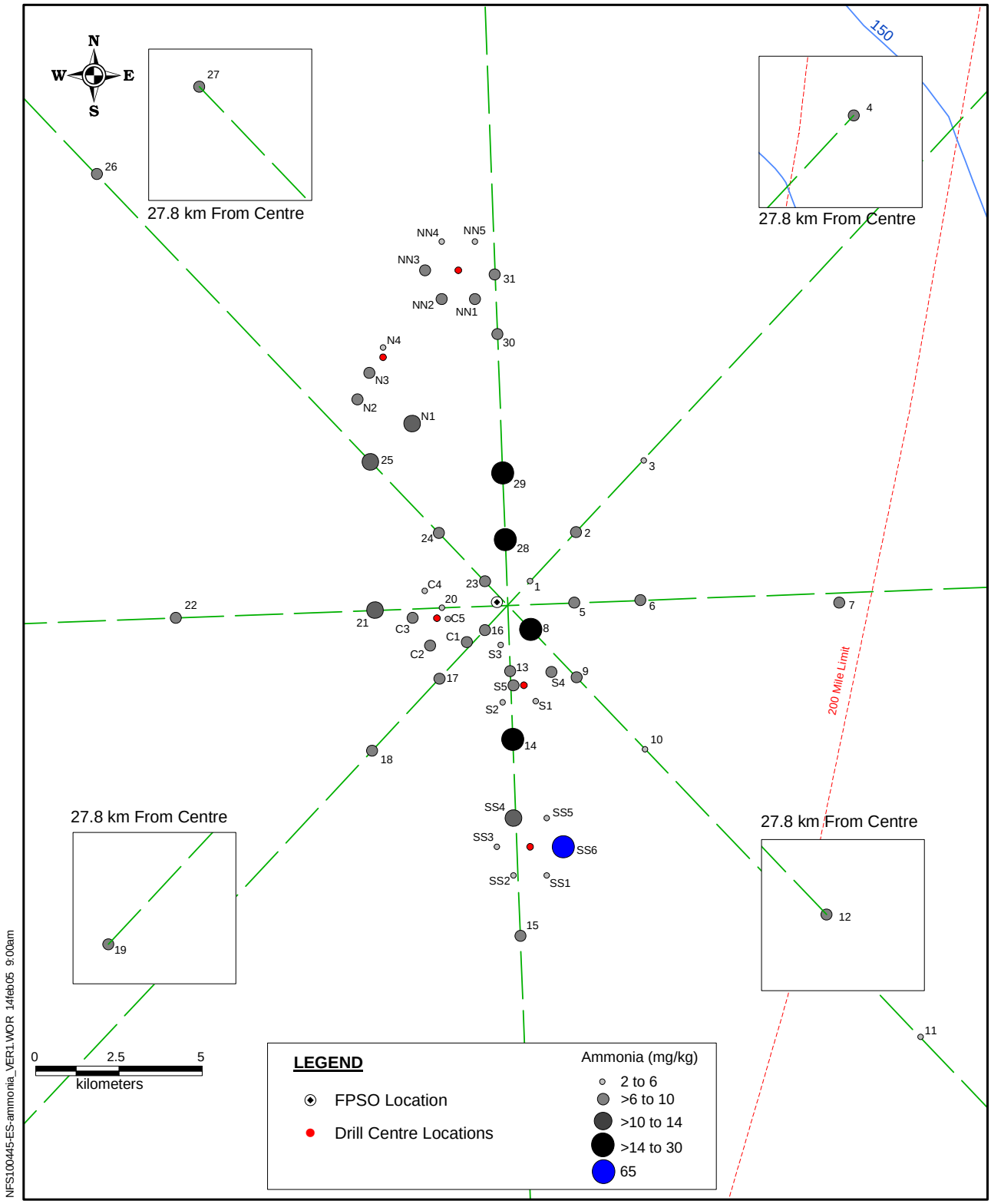


Figure 5-16 Spatial Distribution of Ammonia (2004)

Table 5-14 Results of RM Analyses Comparing Physical and Chemical Characteristics Between 2000 and 2004

Term	df	*F* value for *Y* variables				
		% fines	TOC	Barium	Aluminum	Metals PC1
Full Model						
Among Stations						
Depth	1,32	**20.10*** **	0.51	0.05	1.15	0.01
Northern (N) *d*	1,32	2.30	0.36	4.53*	0.27	0.46
Southern (S) *d*	1,32	4.72*	2.69	**62.47*** **	3.52	6.17*
Central (C) *d*	1,32	0.44	**12.94*** **	7.69**	0.05	1.02
Error 1[1]	32,32	1.50	1.17	1.38	2.24*	1.43
Within Stations						
Year	1,32	3.33	1.25	0.05	0.03	0.00
Year × Depth	1,32	3.68	1.26	0.08	0.03	0.00
Year × N *d*	1,32	0.00	2.21	0.17	1.02	1.15
Year × S *d*	1,32	4.79*	1.44	**85.30*** **	3.62	4.32*
Year × C *d*	1,32	1.28	0.40	**25.04*** **	0.57	0.03
Reduced Model						
Among Stations						
Depth	1,34	**23.25*** **	0.25	0.20	1.00	0.40
Southern (S) *d*	1,34	3.08	0.08	**43.22*** **	5.43*	7.35**
Error 1[1]	34,34	1.55	1.51	1.03	2.17*	1.41
Within Stations						
Year	1,34	6.01*	2.83	2.80	0.00	0.12
Year × Depth	1,34	6.59*	2.98	3.07	0.01	0.14
Year × S *d*	1,34	4.20*	2.55	**40.87*** **	10.05**	8.68**

Notes:
- Appendix B-5 explains terms and tests in the RM regression models
- df = degrees of freedom for numerator (effect), 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* = 37 stations sampled in both 2000 (baseline) and 2004 (EEM)
- All *X* variables and all *Y* variables except Metals PC1 were $\log_{10}$ transformed
- [1] Error 1 = carry-over effects, or persistent differences among stations over time unrelated to depth or distance

$>C_{10}$-C_{21} HCs were not included in the comparisons between years because all values in 2000 were below EQL. Sulphur and ammonia concentrations were not measured in 2000. Aluminum was used as an additional *Y* variable because it is a commonly used reference metal that occurs naturally in marine sediments at high concentrations, and was used in Appendix B-5 (Barium-Aluminum Normalization) for additional analyses of differences in barium concentrations between years.

The full model in Table 5-14 included depth and distances from the Northern, Southern and Central drill centres. The reduced model included depth and distance from the Southern drill centre, which generally had larger effects than distance from the Northern and Central drill centres. The set of 37 stations sampled in both years included only five stations within 5 km of the Northern drill centre, which limited the ability of the RM regression analyses to detect effects from that drill centre. In contrast, the 37 stations included 23 stations within 5 km of the Southern drill centre and 24 stations within 5 km of the Central drill centre, and provided powerful tests of effects from those two drill centres.

Fines content increased significantly with depth in both years (Among Stations Depth term in Table 5-14). The relationship between fines and depth was stronger in 2004 than in 2000 (Figure 5-17), although the Within Stations Year × Depth term was significant only in the reduced model (Table 5-14). Effects of distance from the Northern and Central drill centres were not significant over both years and did not change between years. Changes in the effects of distance from the Southern drill centre (Within Stations Year × Southern *d* effects in Table 5-14) were significant for both the full and reduced RM models. There was no relationship between fines content and distance from the Southern drill centre in 2000, but in 2004, fines content decreased with distance (Figure 5-17). Fines content was also higher at almost every station in 2004 (Figure 5-17). The difference between years varied with depth and with distance from the Southern drill centre (Figure 5-17), which reduced the magnitude (*F*) and the significance of Within Stations Year effects in the RM models (Table 5-14). The change in the relationship between fines content and distance from the Southern drill centre between 2000 and 2004 may be evidence of increased deposition of fines from drilling muds near the Southern drill centre. However, it is unclear why fines content increased in 2004 at every station, and why relationships between fines content and depth changed.

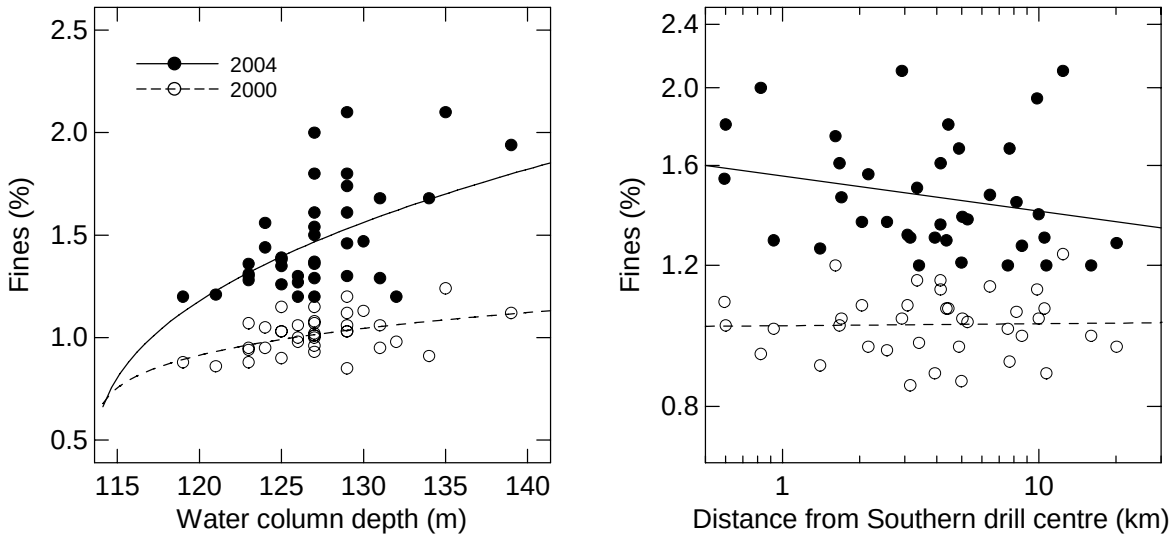


Figure 5-17 Sediment Fines Content Versus Depth and Distance from the Southern Drill Centre (2000 and 2004)

Distance from the Central drill centre significantly affected TOC in both years (Among Stations Central *d* for the full model in Table 5-14). TOC levels were lower near the Central drill centre than at other stations.

The relationship between barium concentrations and distance from the Southern drill centre changed significantly between 2000 and 2004 (Table 5-14; Figure 5-18). In 2000, there was no relationship between barium concentrations and distance. In 2004, concentrations decreased

significantly with distance from the Southern drill centre and concentrations near that drill centre were much higher in 2004 than in 2000. Figure 5-18 confirms results from the larger set of stations sampled in 2004 (Figure 5-9). Background levels of barium were less than 200 mg/kg in time (baseline) as well as space and were approached or met in 2004 at approximately 2 km from drill centres.

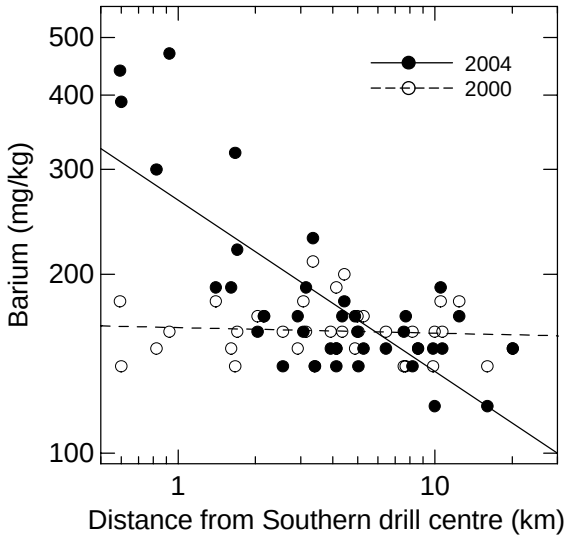


Figure 5-18 Barium Versus Distance from the Southern Drill Centre (2000 and 2004)

The relationship between barium concentrations and distance from the Central drill centre also changed significantly between years (Table 5-14). As noted in the analysis of 2004 data, barium concentrations were greater to the southeast of the Southern drill centre than to the northwest or towards the Central drill centre. That directional difference was not present in 2000, prior to drilling.

In both 2000 and 2004, barium concentrations were higher to the north of the centre of the development than at other stations at similar distances from the centre of the development. In the full RM regression model, the Among Stations effects of distance from the Northern drill centre were significant (Table 5-14), suggesting that some apparent effects of the Northern drill centre on barium concentrations in 2004 may have been partly a function of naturally high concentrations occurring near that centre.

Results of RM regression analyses of Metals PC1 scores and aluminum concentrations, which were highly correlated, were similar (Table 5-14). The relationship between metal concentrations in general, and aluminum concentrations specifically, and distance from the Southern drill centre changed between 2000 and 2004 (Table 5-14; Figure 5-19). In 2000, metal concentrations were unrelated to distance from the Southern drill centre. In 2004, concentrations decreased with

distance from the Southern drill centre. Metal concentrations in 2004 were elevated near the Southern drill centre relative to concentrations at the same stations in 2000, which could be a project effect. However, metal concentrations in 2004 at more remote stations were *lower* than 2000 concentrations at the same stations.

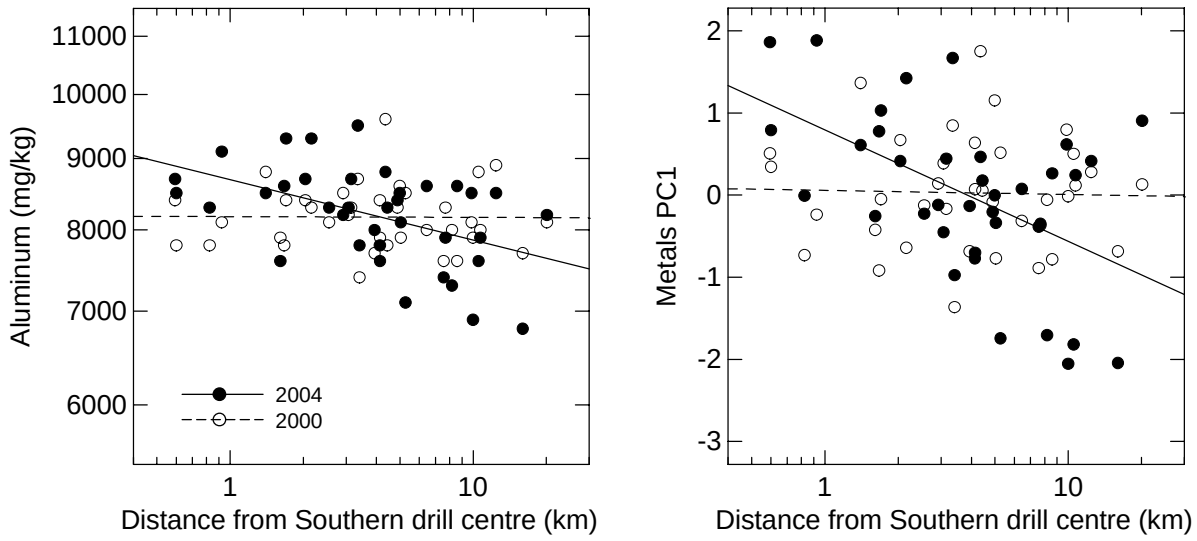


Figure 5-19 Aluminum Versus Metals PC1 Scores and Distance from the Southern Drill Centre (2000 and 2004)

Error 1 or carry-over effects (i.e., persistent differences among stations over time unrelated to depth or distances) were significant only for aluminum (Table 5-14). However, all *F* values for Error 1 were greater than 1, which indicates some added variance attributable to carry-over effects.

5.4.2 Toxicity

In 2004, there were no toxic responses in amphipod tests using Environment Canada's Toxicity Interpretation guidelines (as outlined in section 5.2.2.1). A summary of the amphipod results is provided in Table 5.15. The amphipod laboratory reports are provided in Appendix B-6.

Table 5-15 Amphipod Summary Data and Interpretation

Station	Amphipod Survival (%)	Sample Standard Deviation	Dunnett's t- stat	Statistically Significant	≥ 30% Reduction in Survival	Interpretation (Toxic / Nontoxic)
1	91	10.25	1.939	Yes	No	Nontoxic
2	91	6.52	1.939	No	No	Nontoxic
3	75	14.14	5.451	Yes	No	Nontoxic
4	94	2.24	2.005	No	No	Nontoxic
5	90	3.54	1.649	No	No	Nontoxic
6	94	4.18	0.977	No	No	Nontoxic
7	95	4.08	0.721	No	No	Nontoxic
8	89	8.54	1.880	No	No	Nontoxic
9	94	4.79	0.953	No	No	Nontoxic
10	89	6.29	3.014	Yes	No	Nontoxic
11	99	2.24	1.776	No	No	Nontoxic
12	88	8.37	3.370	Yes	No	Nontoxic
13	88	5	3.349	Yes	No	Nontoxic
14	91	8.54	1.997	No	No	Nontoxic
15	95	4.08	1.481	No	No	Nontoxic
16	84	2.5	3.709	Yes	No	Nontoxic
17	91	6.3	2.344	No	No	Nontoxic
18	93	2.74	1.822	No	No	Nontoxic
19	99	2.24	0.000	No	No	Nontoxic
20	94	4.79	1.654	No	No	Nontoxic
21	88	6.45	3.307	Yes	No	Nontoxic
22	93	5	1.458	No	No	Nontoxic
23	90	4.08	2.282	No	No	Nontoxic
24	93	7.5	1.173	No	No	Nontoxic
25	96	5.48	1.185	No	No	Nontoxic
26	87	8.37	3.380	Yes	No	Nontoxic
27	90	7.07	2.345	No	No	Nontoxic
28	89	4.18	2.431	Yes	No	Nontoxic
29	89	5.48	4.056	Yes	No	Nontoxic
30	92	12.55	0.837	No	No	Nontoxic
31	89	7.42	2.811	Yes	No	Nontoxic
C1	91	4.79	2.315	No	No	Nontoxic
C2	86	7.5	3.638	Yes	No	Nontoxic
C3	95	5.77	1.340	No	No	Nontoxic
C4	91	4.79	3.344	Yes	No	Nontoxic
C5	85	4.08	3.969	Yes	No	Nontoxic
N1	94	7.5	1.956	No	No	Nontoxic
N2	98	5	0.529	No	No	Nontoxic
N3	90	9.35	2.527	Yes	No	Nontoxic
N4	89	6.29	2.675	Yes	No	Nontoxic
NN1	88	4.47	3.095	Yes	No	Nontoxic
NN2	98	4.47	-0.747	No	No	Nontoxic
NN3	93	4.47	0.747	No	No	Nontoxic
NN4	98	5	0.366	No	No	Nontoxic
NN5	96	2.5	1.005	No	No	Nontoxic
S1	79	4.79	4.850	Yes	No	Nontoxic
S2	89	6.29	3.014	Yes	No	Nontoxic
S3	89	6.29	2.976	Yes	No	Nontoxic
S4	93	6.45	1.712	No	No	Nontoxic
S5	74	4.79	7.033	Yes	No	Nontoxic
SS1	92	7.58	2.132	No	No	Nontoxic
SS2	94	4.79	1.476	No	No	Nontoxic

Station	Amphipod Survival (%)	Sample Standard Deviation	Dunnett's t- stat	Statistically Significant	≥ 30% Reduction in Survival	Interpretation (Toxic / Nontoxic)
SS3	93	4.47	1.177	No	No	Nontoxic
SS4	93	8.66	1.835	No	No	Nontoxic
SS5	88	10.41	2.853	Yes	No	Nontoxic
SS6	94	8.22	1.313	No	No	Nontoxic

All Microtox EC50 responses were greater than 197,000 (the highest dilution) in 2004 indicating that there were no toxic responses observed for the Microtox bioassay. Laboratory Reports for the Microtox Bioassay are provided in Appendix B-7.

No toxicity responses were also noted for the 2000 data.

5.4.3 Benthic Community Structure

A total of 25,409 invertebrates were collected from 56 stations in 2004 and a total of 34,481 invertebrates were collected from 46 stations in 2000 (Table 5-16). These totals exclude nemerteans, nematodes, oligochaetes, ostracods and copepods. For both years combined, 86 "families" were collected. Some of the 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, classes or orders) that were not identified to family.

Table 5-16 Taxonomic Composition of Benthic Invertebrate Community Samples (2000 and 2004)

Phylum	Class or Order	No. families (both years)	2004 (EEM) (*n* = 56 stations)		2000 (baseline) (*n* = 46 stations)	
			No. organisms	% of total	No. organisms	% of total
Porifera		1	15	0.1	0	0.0
Cnidaria		5	160	0.6	13	0.0
Annelida	Polychaeta	27	18,907	74.4	26,594	77.1
Mollusca	Bivalvia	15	4,290	16.9	5,857	17.0
	Gastropoda	12	78	0.3	73	0.2
Crustacea	Amphipoda	12	737	2.9	1,184	3.4
	Cirrepedia	1	2	0.0	13	0.0
	Decapoda	1	0	0.0	1	0.0
	Cumacea	3	44	0.2	19	0.1
	Isopoda	4	46	0.2	16	0.0
	Tanaidacea	1	714	2.8	194	0.6
Echinodermata		4	416	1.6	517	1.5
Total		86	25,409		34,481	
Mean no./station			454		616	

In both 2000 and 2004, polychaetes and bivalves accounted for approximately 75% and 17% of the invertebrates collected, respectively (Table 5-16). Thus, these two higher-level taxa accounted for more than 90% of total abundance. Amphipoda, Tanaidacea (2004 only) and Echinodermata were the only other major taxa to account for more than 1% of total abundance. Polychaetes and Bivalves accounted for approximately half of the 86 families collected (i.e., taxonomic richness or diversity). Amphipoda and Gastropoda were represented by 12 families each.

Two dominant polychaete families, Spionidae (mostly *Pionospio steentrupi*) and Paronidae (mostly *Aricidea catherinae*), accounted for more than half of the total number of invertebrates collected in both sample years and occurred at every station (Table 5-17). The bivalve family Tellinidae (mostly *Macoma calcarea*) accounted for greater than 75% of the total number of bivalves collected and occurred at every station in both years. Most other families listed in Table 5-17 were abundant and occurred frequently in both years. However, there were some differences in sub-dominant families between the two years. Specifically, Maldanidae (Polychaeta) and Stenoithidae (Amphipoda) were much more abundant and occurred more frequently in 2004 than in 2000, whereas the reverse was true for Cirratulidae (Polychaeta). Tanaidacea were more abundant in 2004 than in 2000. Cirratulidae were much more abundant in 2000 than in 2004, and Carditidae were relatively abundant in 2000 but were not collected in 2004.

Table 5-17 Dominant Benthic Invertebrate Families (2000 and 2004)

Major Taxon	Family	2004				2000			
		Abundance		Occurrence		Abundance		Occurrence	
		No. organ-isms	% of Total	No. Stations	% of Total	No. organ-isms	% of Total	No. Stations	% of Total
Polychaeta	Spionidae	9,462	37.2	56	100	12,812	37.2	46	100
Polychaeta	Paraonidae	5,004	19.7	56	100	5,020	14.6	46	100
Bivalvia	Tellinidae	3,784	14.9	56	100	4,616	13.4	46	100
Polychaeta	Orbiniidae	1,472	5.8	53	95	1,565	4.5	46	100
Polychaeta	Phyllodocidae	745	2.9	56	100	1,153	3.3	46	100
Tanaidacea		714	2.8	54	96	194	0.6	44	96
Polychaeta	Syllidae	524	2.1	52	93	312	0.9	44	96
Polychaeta	Maldanidae	431	1.7	55	98	405	1.2	46	100
Echinodermata	Echinarachnidae	296	1.2	55	98	348	1.0	46	100
Amphipoda	Dexaminidae	259	1.0	51	91	176	0.5	41	89
Polychaeta	Cirratulidae	257	1.0	32	57	4,412	12.8	46	100
Amphipoda	Haustoriidae	227	0.9	50	89	641	1.9	46	100
Bivalvia	Carditidae	0	0.0	0	0	441	1.3	41	89
Total		23,175	91.2			32,095	93.1		

These differences between years did not appear to be taxonomic artifacts. In each year, all but one cirratulid was identified as *Chaetozone* serosa. *Praxillella praetermissa* was the dominant Maldanidae taxon and *Guernea nordenskioldi* was the dominant Stenoithidae taxon in both years. Tanaidacea were not identified to lower taxonomic levels by either taxonomist, and would be identifiable to order by any taxonomist. The complete absence of Carditidae (all *Cyclocardia* spp. in 2000) in 2004 was more surprising, but the taxonomist used in 2004 was capable of recognizing and identifying these organisms at the family level or lower, and they were not erroneously included in another family.

5.4.3.1 Preliminary Analysis

Non-Metric Multi-dimensional Scaling

Figure 5-20 provides the 2-dimensional NMDS plot for the combined analysis of community composition for 56 stations sampled in 2004 and 46 stations sampled in 2000. The NMDS was based on relative abundances of families. The stress coefficient was 0.17, which represents a reasonable fit to the original pair-wise similarity matrix.

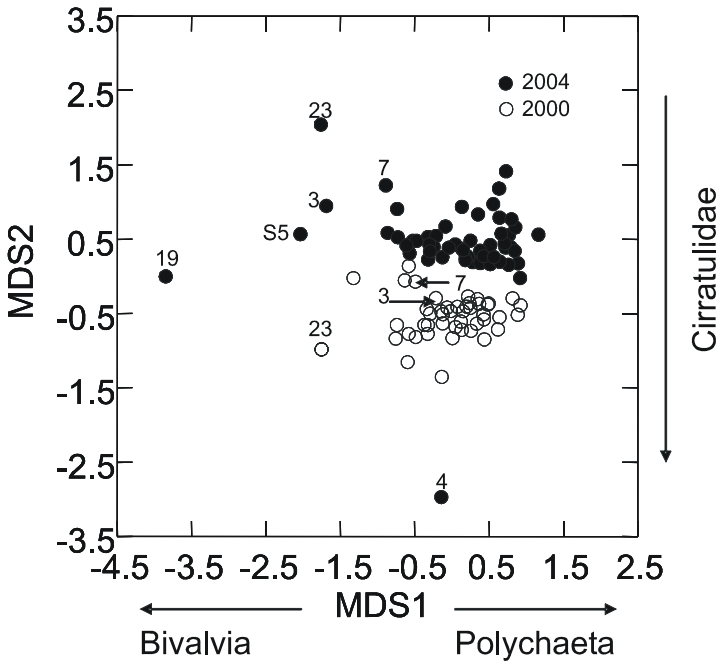


Figure 5-20 NMDS Plot Based on Relative Abundances of Invertebrate Taxa (2000 and 2004)

MDS1 scores were positively correlated with the relative abundance of polychaetes and negatively correlated with the relative abundance of bivalves. Polychaetes and bivalves accounted for more than 90% of the total number of organisms collected. The relative abundances of the two taxa should be negatively correlated (i.e., relative abundance of one taxon is 90-100% minus the relative abundance of the other), and the primary axis of community variance (MDS1 scores) should reflect that polcyhaete:bivalve contrast.

MDS1 scores (polychaete:bivalve abundances) varied mostly among stations within years. MDS2 scores clearly separated the two sample years, with scores lower in 2000 than in 2004. MDS2 scores were negatively correlated with the relative abundance of Cirratulidae and to a lesser extent Carditidae, which were more abundant and common in 2000 than in 2004 (Table 5-17). MDS2 scores were also positively correlated with relative abundances of several of the dominant families in Table 5-17 (e.g., Paraonidae, Tellinidae, Syllidae). With few Cirratulidae and no Carditidae collected in 2004, the relative abundances of other dominant taxa must increase so that relative abundances sum to 100%.

Summary Statistics

Table 5-18 provides summary statistics for the seven benthic invertebrate community measures. Total abundance per station was lower in 2004 than in 2000 (see also Table 5-16). Standing crop, richness and evenness were similar between years, and richness and diversity were slightly lower in 2004. With the exception of a few extreme stations and values in 2004, MDS1 scores were similar between years, and MDS2 scores were greater in 2004 than in 2000 (see also Figure 5-20).

Table 5-18 Summary Statistics for Benthic Invertebrate Community Summary Measures (2000 and 2004)

Variable	Minimum	Maximum	Median	Mean	SD
2004 (*n* = 56 stations)					
Total abundance	262	847	433	454	126
Standing crop	36	351	153	163	70
Richness	21	34	26	27	3
Diversity	3.2	6.1	4.1	4.3	0.7
Evenness	0.12	0.21	0.16	0.16	0.02
MDS1	–3.84	1.17	0.20	0.03	0.87
MDS2	–2.97	2.04	0.42	0.44	0.59
2000 (*n* = 46 stations)					
Total abundance	322	1,198	717	750	226
Standing crop	56	257	145	151	47
Richness	19	37	29	29	4
Diversity	3.2	7.5	5.0	4.9	0.9
Evenness	0.12	0.31	0.16	0.17	0.04
MDS1	–1.75	0.92	0.03	–0.03	0.54
MDS2	–1.35	0.14	–0.51	–0.54	0.28

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

Table 5-19 provides summary statistics for relative (%) abundances of the five major taxa. Minima for polychaete abundances were approximately 50%, indicating that in both years half or more of the invertebrates collected at every station were polychaetes.

Table 5-19 Summary Statistics for Relative Abundances of Major Taxa (2000 and 2004)

Variable	Minimum	Maximum	Median	Mean	SD
2004 (*n* = 56 stations)					
Polychaeta	49.6	86.9	75.2	73.9	8.8
Bivalvia	5.6	42.0	15.7	17.2	8.0
Amphipoda	0.0	7.6	2.5	2.9	1.6
Tanaidacea	0.0	10.1	2.4	2.9	1.8
Echinodermata	0.0	4.7	1.6	1.7	1.0
2000 (*n* = 46 stations)					
Polychaeta	53.9	88.7	77.6	76.7	6.8
Bivalvia	7.0	41.5	16.3	17.1	6.3
Amphipoda	0.8	8.0	3.2	3.6	1.6
Tanaidacea	0.0	2.3	0.5	0.6	0.4
Echinodermata	0.6	5.3	1.4	1.6	0.9

Note: - Values are % of total abundance

5.4.3.2 Correlations Within and Among Groups of Variables (2004)

Correlations among invertebrate community summary measures and between those summary measures and major taxon relative abundances for 2004 samples are provided in Table 5-20. Most of these correlations were expected to be non-zero and the primary objective in this analysis was to identify groups of similar variables for which results and any project effects could be expected to be similar. For example, diversity (*D*) must be positively correlated with its two components, richness and evenness ($D=S\times E$). Because of that mathematical relationship, correlations between diversity and other variables were approximately equal to the sum of correlations between those variables, and richness and evenness. Correlations for 2000 were generally similar in sign and often magnitude (Husky Energy 2001), again an indication that non-zero correlations can be expected in the absence of drilling effects.

Table 5-20 Spearman Rank Correlations (r_s) Among Benthic Invertebrate Community Summary Measures and Between those Measures and Relative Abundances of Major Taxa (2004)

	Total Abundance	Standing Crop	Richness	Diversity	Evenness	MDS1	MDS2
Summary Measures							
Standing crop	–0.235						
Richness	0.367**	0.113					
Diversity	–0.211	0.152	0.359**				
Evenness	**–0.473*** **	0.044	–0.315*	**0.732*** **			
MDS1	0.158	–0.168	–0.343*	**–0.666*** **	–0.406**		
MDS2	0.345*	0.039	0.205	–0.084	–0.222	–0.150	
Relative Abundances of Major Taxa							
Polychaeta	0.307*	–0.293*	–0.327*	**–0.539*** **	–0.291*	**0.753*** **	–0.099
Bivalvia	–0.200	0.229	0.261	0.427**	0.216	**–0.728*** **	0.157
Amphipoda	0.056	–0.121	0.282*	0.266*	0.107	–0.037	–0.049
Tanaidacea	–0.270*	0.112	0.278*	0.430**	0.242	–0.256	–0.132
Echinodermata	–0.249	**0.549*** **	0.018	0.140	0.151	–0.056	0.115

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

Total abundance was positively correlated with richness (*S*), negatively correlated with evenness (E) or *D*/*S*. Consequently, the correlation between abundance and diversity was weak (near zero) and not significant. Abundance and richness usually will be positively correlated, with more taxa collected when more individuals are collected. As expected, total abundance was positively correlated with the relative abundance of polychaetes, the most abundant major taxon. More specifically, total abundance was positively correlated with absolute and relative abundances of Paraonidae, which partially accounts for the positive correlation between total abundance and MDS2 scores. MDS2 scores were positively correlated with relative abundances of Paraonidae, but also some other dominant polychaete and non-polychaete families.

Standing crop was not strongly correlated with any variable except the relative abundance of echinoderms (Table 5-20). Larger organisms such as echinoderms would account for most of the standing crop or wet weight per station.

Diversity was more strongly positively correlated with evenness than with richness (Table 5-20). Therefore, evenness rather than richness was the major component affecting diversity values. Richness, evenness and especially diversity were all negatively correlated with MDS1 and the relative abundance of Polychaeta, and more weakly positively correlated with relative abundances of Bivalvia, Amphipoda and Tanaidacea.

MDS1 and relative abundance of polychaetes are both measures of polychaete dominance, and were strongly positively correlated (Table 5-20; see also Figure 5-20). Bivalvia, Amphipoda and Tanaidacea can be considered “non-polychaetes” or 100 minus % polychaetes, with relative abundances greater where polychaetes were less dominant. Tanaidacea is a single “family” (actually order) and most bivalves also belonged to a single family (Tellinidae). Increased abundances of single taxa such as Tanaidacea and Tellinidae would normally decrease richness, diversity and evenness. However:

- where Tanaidacea and Tellinidae were more abundant, other taxa or families (including sub-dominant polychaetes) were more likely to be collected, although usually in limited numbers
- where polychaetes were most dominant, only a few taxa or families (e.g., Spionidae and Paraonidae) were abundant and accounted for that dominance

As noted elsewhere (e.g., Figure 5-20), MDS1 scores were positively correlated with relative abundances of polychaetes and negatively correlated with relative abundances of bivalves. MDS2 scores were uncorrelated with the relative abundances of major taxa, because the scores were mostly a function of differences within Polychaeta (e.g., Cirratulidae versus Paraonidae and several other dominants) and between years.

5.4.3.3 Distance and Depth Effects (2004)

Extreme and Excluded Stations

Reference stations 4 and 19 represented extreme depth values that substantially affected regressions on depth, and were also outliers in terms of community composition and MDS scores (Table 5-21, Figure 5-20). MDS1 scores and abundances of polychaetes relative to bivalves were lower at station 19 than elsewhere. MDS2 scores were much lower at station 4 than at other stations in 2004 and also in 2000. Station 4 accounted for 70% of the Cirratulidae collected from 56 stations in 2004, and the relative abundance of Cirratulidae was greater there than at any other station sampled in either year. Station S5, the most contaminated station, was not an outlier in any regression, although total abundance and MDS1 scores there were lower than at most or all stations.

Table 5-21 Benthic Invertebrate Community Variables for All Stations, the Trimmed Set of Stations and Extreme (Near and Far) Stations

Data set	Statistic/ Station	Distance[1] (km)	Depth (m)	*Y* variable						
				Abundance	Standing crop	Richness	Diversity	Evenness	MDS1	MDS2
All stations (*n* = 56)	Minimum	0.30	108	262	36	21	3.2	0.12	–3.84	–2.97
	Maximum	26.19	175	847	351	34	6.1	0.21	1.17	2.04
	Median	3.08	127	433	153	26	4.1	0.16	0.20	0.42
Trimmed (*n* = 50)	Minimum	0.33	119	291	**36**	**21**	**3.2**	**0.12**	–1.76	–0.02
	Maximum	16.00	139	**847**	**351**	**34**	**6.1**	**0.21**	**1.17**	**2.04**
	Median	2.97	127	437	153	26	4.1	0.16	0.23	0.52
Near Northern	*N4*	**0.30**	126	368	206	28	5.2	0.19	–0.52	0.48
	N3	0.63	126	328	152	25	3.9	0.16	0.56	0.26
Near Southern	*S5*	0.31	127	**262**	251	27	4.1	0.15	–2.03	0.57
	13	0.59	127	379	246	26	3.6	0.14	0.25	0.48
	S1	0.60	127	429	109	24	3.8	0.16	0.92	–0.02
Near Central	C5	0.33	124	404	148	23	4.3	0.19	0.19	0.24
	20	0.35	127	368	160	26	4.5	0.17	0.38	0.18
	C3	0.74	123	476	198	28	3.8	0.14	0.71	0.44
References	*4 (NE)*	**26.19**	**175**	586	112	30	5.8	0.19	–0.14	**–2.97**
	12 (SE)	25.85	137	549	120	25	3.7	0.15	0.77	0.55
	19 (SW)	26.18	**108**	298	181	23	4.9	0.21	**–3.84**	–0.00
	27 (NW)	20.03	123	389	93	24	3.5	0.15	0.76	0.15

Notes:
- Italics indicate stations that were trimmed (N4, S5 and Reference stations)
- Bold indicates minima or maxima for all 56 stations that occurred either in the trimmed data set or at the extreme stations
- [1] Distance to nearest active drill centre (Northern, Southern, Central)
- Richness, diversity, evenness and MDS scores are based families

Regression Analyses on Summary Measures

Tables 5-22 and 5-23 provide regression results for the full and trimmed data sets. Overall, there was little evidence of distance effects on summary measures but there were some strong relationships with depth, and some strong effects of extreme values.

Table 5-22 Results of Regressions of Benthic Invertebrate Community Variables on Depth and Distances from Active Drill Centres for All Stations (2004)

Model/term	Result	*Y* variable						
		Abundance	Standing crop	Richness	Diversity	Evenness	MDS1	MDS2
X = Depth + distances (*d*) from all drill centres								
Overall	r^2	0.156	0.040	0.194*	0.179*	0.021	0.049	0.235**
Depth	Slope *(b)*	1.117	–303	45.1**	11.11**	0.111	2.329	**–11.418*** **
Northern *d*	Slope *(b)*	0.057	–29	–2.1	–0.32	0.002	0.015	–0.129
Southern *d*	Slope *(b)*	0.092*	–12	0.6	–0.04	–0.004	0.152	–0.209
Central *d*	Slope *(b)*	–0.056	27	–0.8	0.00	0.005	–0.543	0.227
X = Depth + distances (*d*) from Northern and Southern drill centres								
Overall	r^2	0.131	0.025	0.187*	0.179*	0.018	0.010	0.220**
Depth	Slope *(b)*	0.960	–229	42.8*	11.11**	0.123	0.813	**–10.786*** **
Northern *d*	Slope *(b)*	0.046	–24	–2.3*	–0.32	0.003	–0.095	–0.084
Southern *d*	Slope *(b)*	0.056	5	0.12	–0.04	–0.001	–0.192	–0.066
X = Distance from nearest drill centre								
Overall	r^2	0.076*	0.001	0.000	0.004	0.004	0.020	0.008
Distance	Slope *(b)*	0.064*	–5	0.0	0.09	0.003	–0.254	–0.110
X = Distance from nearest of Northern and Southern drill centres								
Overall	r^2	0.104*	0.006	0.001	0.000	0.000	0.004	0.017
Distance	Slope *(b)*	0.085*	–13	–0.2	–0.03	0.001	–0.121	–0.180

Notes:
- *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$
- For regressions with a single *X* (distance) variable, *p* are the same for r^2 and slopes (*b*)
- All *X* variables and abundance were $\log_{10}$ transformed
- Richness, diversity, evenness and MDS scores are based families

Table 5-23 Results of Regressions of Benthic Invertebrate Community Variables on Depth and Distance from Active Drill Centres for the Trimmed Data Set of 50 Stations (2004)

Model/term	Result	*Y* variable						
		Abundance	Standing crop	Richness	Diversity	Evenness	MDS1	MDS2
X = Depth + distances (*d*) from all drill centres								
Overall	r^2	0.116	0.045	0.184	0.291**	0.112	**0.474*** **	0.210*
Depth	Slope *(b)*	0.579	–349	86.8*	**29.86*** **	0.611*	**–36.647*** **	5.303
Northern *d*	Slope *(b)*	0.103	–5	–1.8	–0.209	0.005	–0.076	0.438*
Southern *d*	Slope *(b)*	0.107	26	1.3	–0.113	–0.011	–0.173	0.244
Central *d*	Slope *(b)*	–0.025	21	–1.4	–0.284	–0.003	0.293	0.093
X = Depth + distances (*d*) from Northern and Southern drill centres								
Overall	r^2	0.111	0.037	0.166*	0.274**	0.110	**0.455*** **	0.204*
Depth	Slope *(b)*	0.332	–145	73.1*	**27.07*** **	0.580*	**–33.771*** **	6.213
Northern *d*	Slope *(b)*	0.097	1	–2.1	–0.277	0.004	–0.005	0.460*
Southern *d*	Slope *(b)*	0.091	39	0.5	–0.284	–0.013	0.003	0.300
X = Distance from nearest of Northern and Southern drill centres								
Overall	r^2	0.101*	0.022	0.006	0.000	0.004	0.021	0.119*
Distance	Slope *(b)*	0.110*	27	0.8	–0.04	–0.005	–0.298	0.410*
X = Distance from Southern drill centre								
Overall	r^2	0.047	0.014	0.047	0.001	0.017	0.031	0.044
Distance	Slope *(b)*	0.064	27	1.9	0.06	–0.008	–0.309	0.213

Notes:
- Trimmed data set excludes four Reference stations (4, 12, 19, 27) and N4 and S5 (see Table 5-21 for values for excluded stations)
- *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$ (in bold)
- For regressions with a single *X* (distance) variable, *p* are the same for r^2 and slopes (*b*)
- All *X* variables and abundance were $\log_{10}$ transformed
- Richness, diversity, evenness and MDS scores are based families

The only consistent significant distance effects occurred for total abundance, which generally increased with distance from the Southern and Northern drill centres (Tables 5-22 and 5-23; Figure 5-21). Abundances within 2 km of the two drill centres were all below 500 organisms/station (Figure 5.21). Beyond 2 km, both high (more than 500 organisms/station) and low abundances occurred, and there were no apparent distance or directional gradients (Figures 5-21 and 5-22).

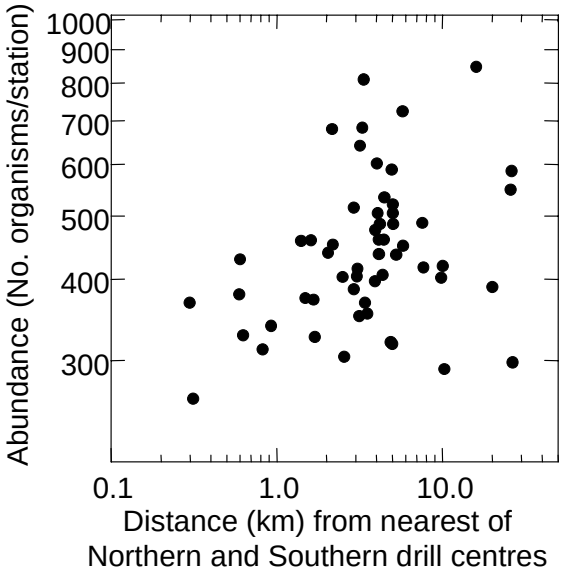


Figure 5-21 Abundance Versus Distance from Drill Centres (2004)

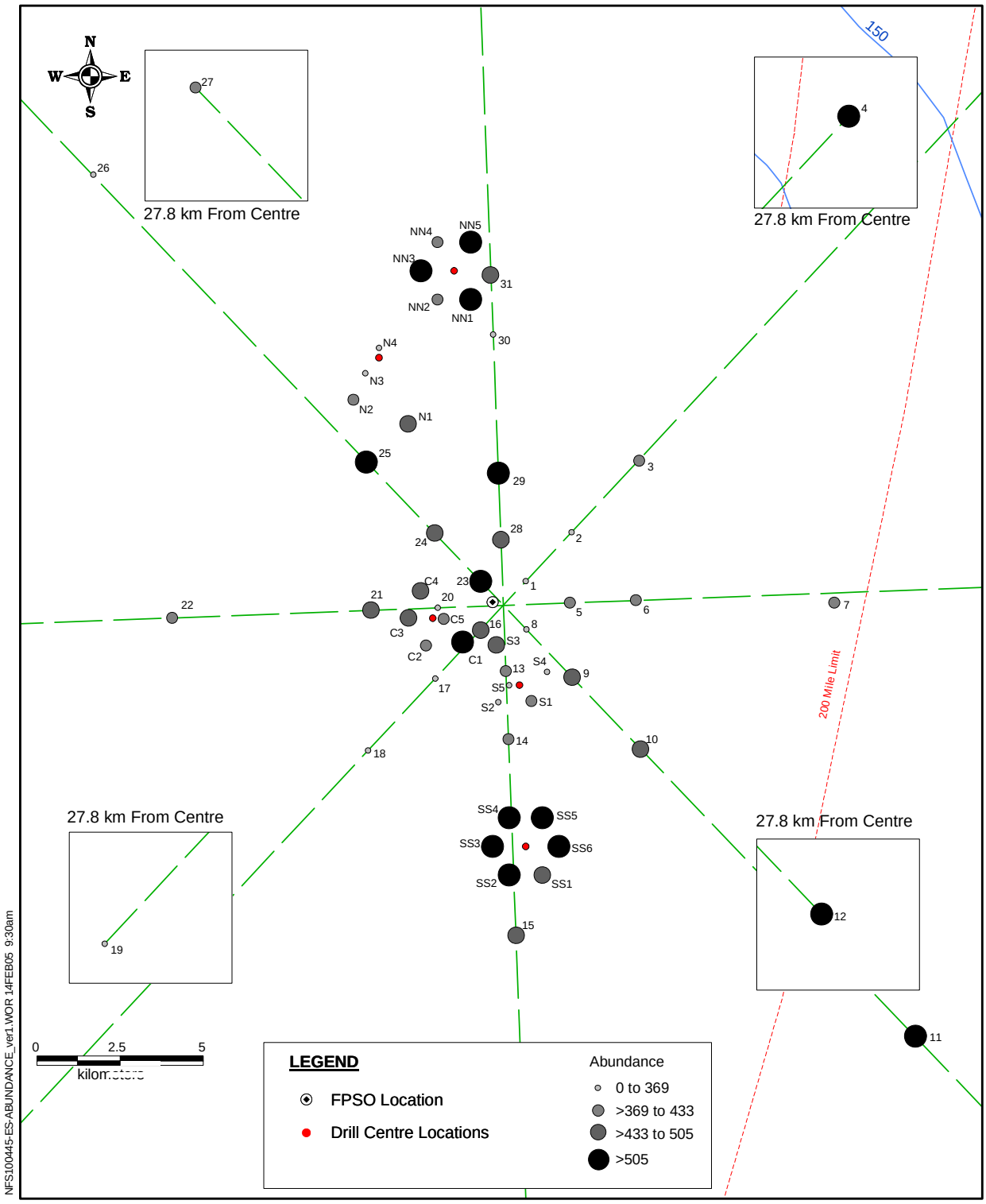


Figure 5-22 Spatial Distribution of Abundance (2004)

Depth and distance effects on standing crop were never significant (Table 5-22 and 5-23).

Diversity increased significantly with depth, and that increase was significant at $p \leq 0.001$ for the trimmed data (Table 5-23; Figure 5-23). The depth effects were less significant for the full data set (compare Table 5-22 and 5-23). Figure 5-24 provides the spatial distribution of diversity, indicating that values increased to the northeast when strong depth effects were present. Richness and evenness also increased with depth, although those relationships were not always significant at $p \leq 0.05$ and were weaker relationships between diversity and depth (Tables 5-22 and 5-23).

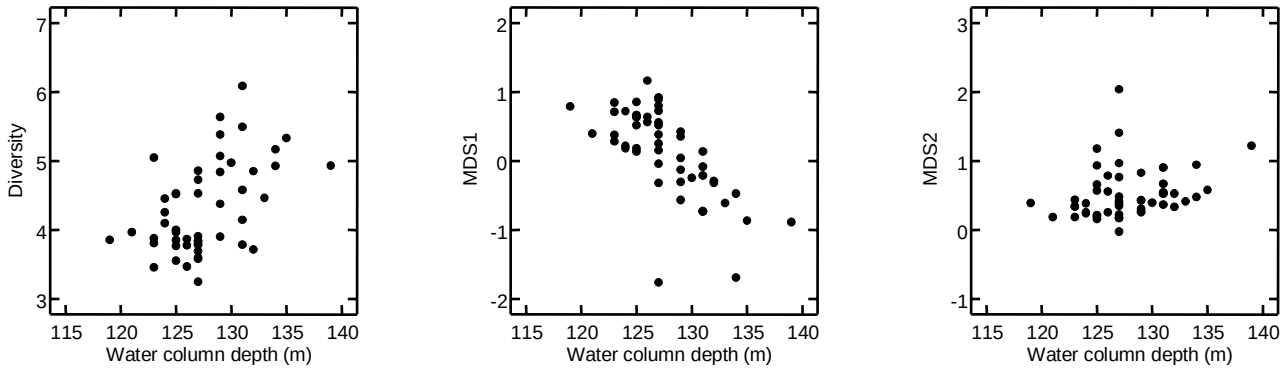


Figure 5-23 Diversity and MDS Scores Versus Depth (2004)

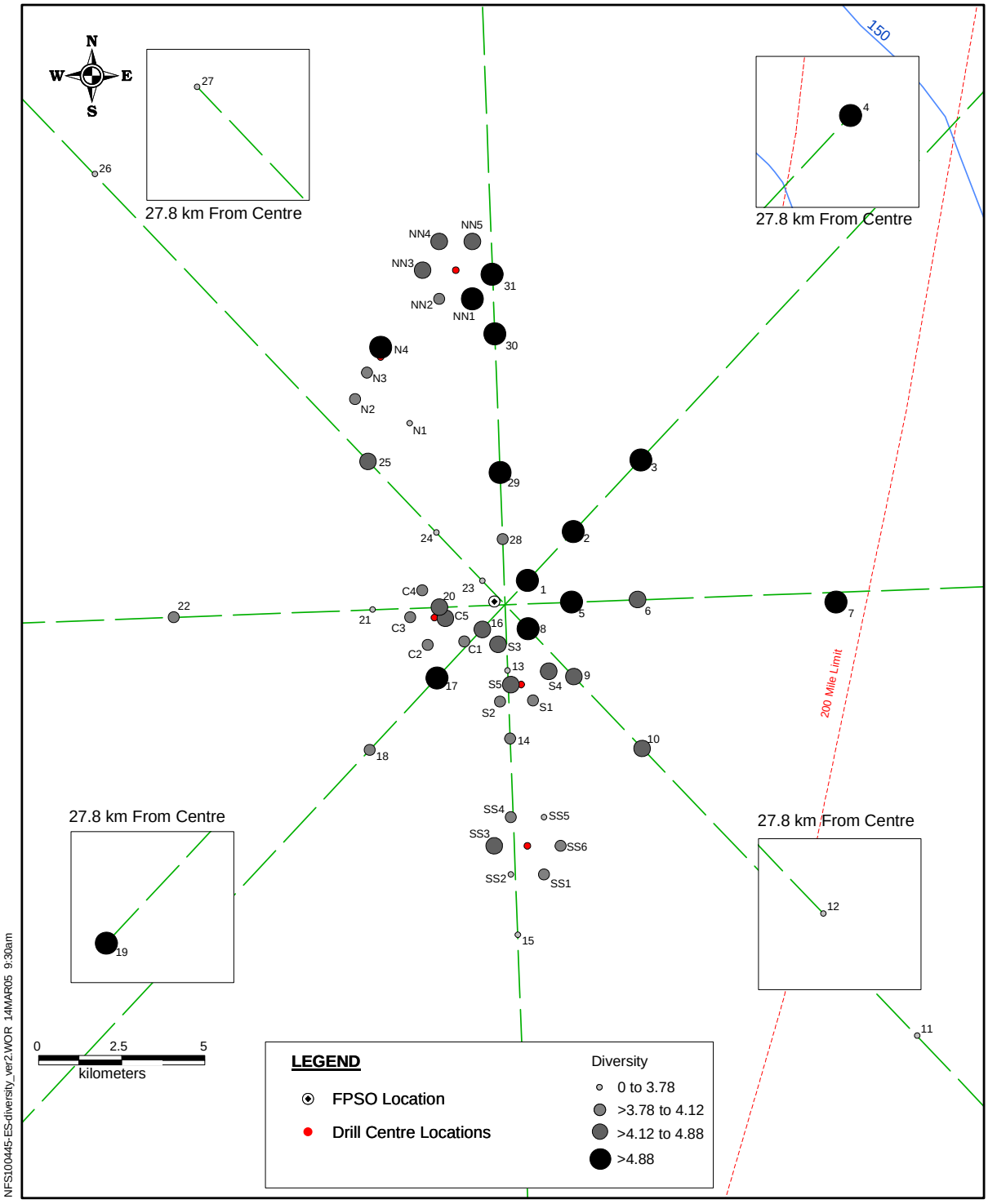


Figure 5-24 Spatial Distribution of Diversity (2004)

Decreases in MDS1 scores (polychaetes:bivalves) with depth were highly significant ($p \leq 0.001$ for the trimmed data set) (Table 5-23; Figure 5-23). However, there was no decrease with depth for all stations (Table 5-22), because MDS1 scores were much lower at station 19 (shallowest depth) than at any other station (Table 5-21). The depth relationship for all stations was effectively a two-point regression between station 19 (lowest MDS1 score, shallowest depth) and station 4 (intermediate MDS1 score, greatest depth), the opposite of the relationship shown in Figure 5-23 for other stations.

A similar reversal of depth relationships with MDS2 scores also occurred for all stations versus the trimmed data set. For the trimmed data set (i.e., at most stations and intermediate depths), MDS2 scores increased significantly although weakly ($0.01 < p < 0.05$) with depth (Table 5-23; Figure 5-23). For all stations, there was a highly significant ($p \leq 0.001$) *decrease* with depth, which was effectively a two-point regression between stations 4 (extreme low MDS2 score; Table 5-21) and 19.

For the trimmed data set, MDS2 scores also increased significantly with distance from the Northern and Southern drill centres (Table 5-23). That distance effect should not be regarded as evidence of drilling effects. Higher MDS2 scores occurred mostly at intermediate distances (5-10 km), not at stations near either the Northern or Southern drill centres (Figure 5-25). Any distance effects on MDS2 scores for the trimmed data set were trivial compared to the difference between station 4 and all other stations (Figure 5-25), and the effects of distances from the Northern and Southern drill centres were negative not positive for all stations (Table 5-22).

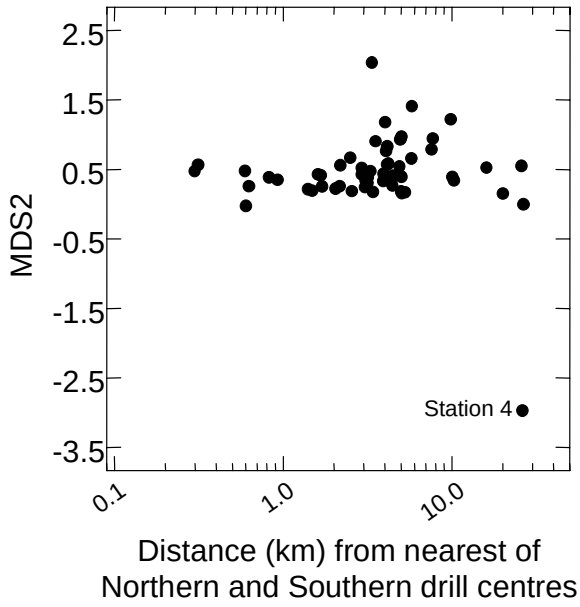


Figure 5-25 MDS2 Scores Versus Distance from Nearest of Northern and Southern Drill Centres

Correlation Analyses on Relative Abundances of Major Taxa

Rank correlations between the relative abundances of the five major taxa, depth and distances are provided in Table 5-24. The relative abundance of polychaetes decreased with depth and the relative abundance of bivalves increased with depth. These results are consistent with regression results for MDS1 scores (polychaetes:bivalves) for the trimmed data set (Table 5-23). The extreme MDS1/polychaete:bivalve and depth values at stations 4 and 19 had minimal influence on correlations between ranks of relative abundances of the two dominant taxa and depth, since ranks remove most of the differences between those extremes and other values. Correlations between relative abundances of these two dominant taxa and distance measures were weak. The relative abundance of Tanaidacea decreased significantly with distance from the Northern but not from the Southern drill centre; whereas the relative abundance of Amphipoda increased significantly with distance from the Southern but not from the Northern drill centre. There were no depth or distance effects on the relative abundance of echinoderms.

Table 5-24 Spearman Rank Correlations (r_s) Between Relative Abundances of Major Taxa and Depth and Distances from Drill Centres (2004)

X variable	Relative abundances (*Y*)				
	Polychaeta	**Bivalvia**	**Amphipoda**	**Tanaidacea**	**Echino-dermata**
Water column depth	**–0.462*****	**0.569*****	0.065	–0.049	0.066
Distance from:					
Northern drill centre	0.294*	–0.238	–0.173	**–0.495*****	0.154
Southern drill centre	–0.071	–0.022	0.392**	0.186	–0.162
Central drill centre	–0.172	0.129	0.141	0.004	0.024
Nearest of Northern and Southern	0.208	–0.227	0.316*	–0.146	–0.102

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

5.4.3.4 Comparison Between Years (2000 and 2004)

Summary Community Measures

Results of RM regression analyses comparing benthic invertebrate community summary measures between 2000 and 2004 are provided in Table 5-25. As for physical and chemical variables, a full model with depth and distances from all three drill centres as *X* variables, and a reduced model with only depth and distance from the Southern drill centre as *X* variables, were tested.

Table 5-25 Results of Repeated Measures (RM) Regression Analyses Comparing Benthic Invertebrate Community Summary Measures Between 2000 and 2004

Term	df	*F* values for *Y* variables						
		Total Abundance	**Standing crop**	**Richness**	**Diversity**	**Evenness**	**MDS1**	**MDS2**
Full Model								
Among Stations								
Depth	1,32	**12.46*** **	0.85	8.23**	**13.69*** **	0.49	**13.20*** **	6.36*
Northern (N) *d*	1,32	2.65	0.68	3.92	6.99*	0.17	0.15	0.99
Southern (S) *d*	1,32	0.58	1.52	0.01	0.25	0.27	1.30	1.40
Central (C) *d*	1,32	5.34*	1.81	4.57*	4.40*	0.01	2.27	0.14
Error 1[1]	32,32	0.54	1.60	1.63	0.60	0.65	2.79**	0.76
Within Stations								
Year	1,32	2.41	0.35	0.24	0.99	1.11	10.12**	0.75
Year × Depth	1,32	2.65	0.34	0.31	0.8	1.07	9.77**	0.84
Year × N *d*	1,32	0.02	0.01	2.90	3.46	0.52	1.97	0.58
Year × S *d*	1,32	8.06**	0.01	1.71	0.80	2.01	0.69	1.05
Year × C *d*	1,32	0.02	0.22	0.01	0.54	0.29	1.41	0.80
Reduced Model								
Among Stations								
Depth	1,34	8.88**	0.09	2.88	5.41*	0.56	11.50**	9.67**
Southern (S) *d*	1,34	7.17*	4.07	0.17	0.05	0.76	0.14	1.85
Error 1[1]	34,34	0.66	1.73	1.95*	0.74	0.64	2.67**	0.76
Within Stations								
Year	1,34	2.74	0.23	0.09	2.60	2.23	**16.26*** **	0.50
Year × Depth	1,34	3.04	0.23	0.10	2.57	2.25	**16.20*** **	0.61
Year × S *d*	1,34	**13.28*** **	0.06	0.91	1.59	2.90	0.61	0.23

Notes:
- Appendix B-5 explains terms and tests in the RM regression models
- df = degrees of freedom for numerator (effect), 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* = 37 stations sampled in both 2000 and 2004
- All *X* variables and abundance were $\log_{10}$ transformed
- [1] Error 1 = carry-over effects, or persistent differences among stations over time unrelated to depth or distance
- Richness, diversity, evenness and MDS scores are based families

Among Stations Depth terms were significant for total abundance for both the full and reduced RM models, and the Year × Depth terms were not significant (Table 5-25). These results indicate there was a common depth relationship in both years, which was somewhat surprising. Analysis of the full set of stations sampled in 2004 indicated there were no depth effects on total abundance (Tables 5-22 and 5-23), which appeared to be true for the subset of 37 stations used for the RM regression analyses (Figure 5-26). In contrast, abundance increased with depth in 2000 for the 37 RM stations (Figure 5-26), although not for the larger set of all stations sampled that year (Husky Energy 2001).

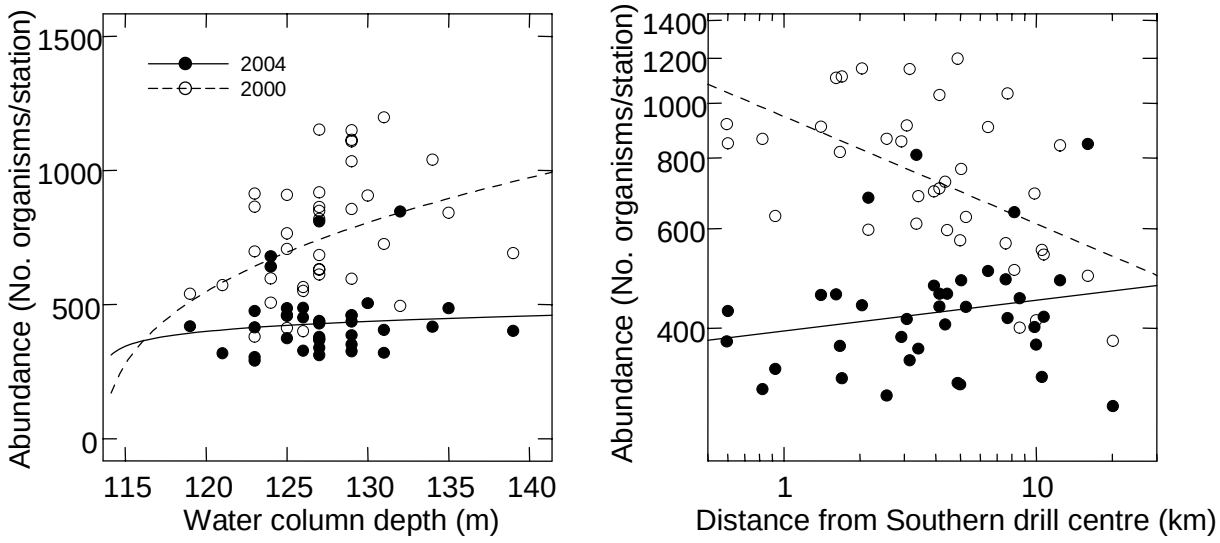


Figure 5-26 Abundance Versus Depth and Distance from the Southern Drill Centre (2000 and 2004)

Total abundance increased with distance from the Southern drill centre in 2004, but decreased with distance in 2000 (Figure 5-26). Thus, the Year × Southern *d* terms were significant in both the full and reduced RM regression models (Table 5-25). There was no change from 2000 to 2004 in the relationship between total abundance and distance from the Northern drill centre. Therefore, the gradient appears to be natural. Abundances were greater in 2000 than in 2004 (Figure 5-26). However, the Year effect was not significant in the RM models because the differences between years varied with distance from the Southern drill centre (i.e., were much greater near that drill centre than at more remote stations).

None of the terms in the RM models were significant for standing crop (Table 5-25), which differed little between years (Table 5-18).

Diversity increased significantly with depth in both years (Among Stations Depth term in Table 5-25), as it did for the full set of stations sampled in 2004 (Tables 5-22 and 5-23). The significant Among Stations effects for distance from the Central drill centre in the full RM model (Table 5-25) can be regarded as directional "correctors" for the overall depth relationship. With those terms dropped in the reduced model, depth effects were less significant. Results for richness in the RM models were similar to results for diversity. Evenness was unaffected by depth and distance and did not differ between Years (Table 5-25; see also Table 5-18).

Distance effects on MDS scores were never significant over both years, and never differed in slope or intercept between years (Table 5-25). MDS1 scores (polychaetes:bivalves) decreased

significantly with depth in both years (significant Among Stations effects in Table 5-25). That depth relationship was stronger in 2004 than in 2000 (Within Stations Year × Depth effects for both models in Table 5-25; Figure 5-27)

Relationships between MDS2 scores and depth were more remarkable. In both years, MDS2 scores increased with depth (significant Among Stations effects in Table 5-25; Figure 5-27). MDS2 scores (reduced abundances of Cirratulidae and absence of Carditidae) were higher in 2004 at every one of the same 37 stations sampled in 2000 (Figure 5-27). However, despite that obvious difference between years, Within Station Year effects were not significant (Table 5-25). That is a statistical artifact, which was not specific to the RM model or the log transformation used for depth. The RM model was unable to distinguish between differences in intercepts (Year) versus slopes (Year × Depth) for the MDS2-depth regressions, although the difference in depth slopes seems minor in Figure 5-27. The same results (failure to distinguish between Year effects on intercepts versus slopes, with neither significant) were obtained with analyses of untransformed depths, and with ANCOVA comparing depth regressions between years for the 37 stations sampled in both 2004 and 2000 or the entire set of 102 stations sampled in either year. At the same time, any analysis that excluded depth, or simultaneously tested for a difference in *either* depth intercepts or slopes, readily identified a difference between years, usually at $p \leq 0.001$. Otherwise, there were no common effects of distances from the drill centres, and no differences in distance relationships between years, which are more relevant results for impact assessment.

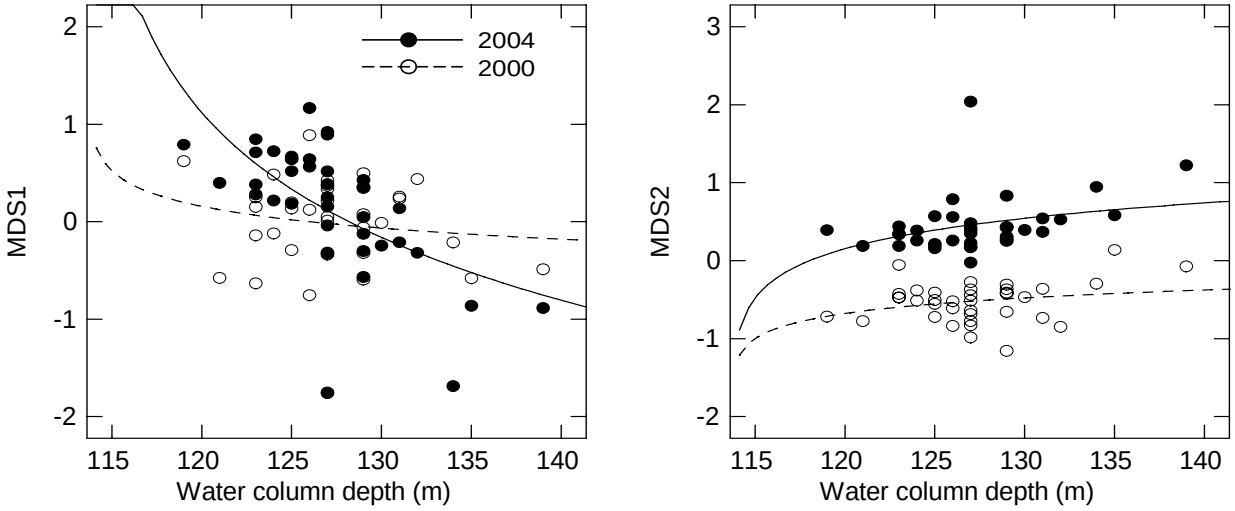


Figure 5-27 MDS Scores Versus Depth (2000 and 2004)

Error 1 or carry-over effects were significant for MDS1 scores in both the full and reduced RM models (Table 5-25). Figure 5-20 provides evidence of carry-over effects for MDS1 for stations 3, 7 and 23, which extended to a lesser degree to other stations sampled in both years. Within each year, MDS1 scores and polychaete:bivalve abundances were lower at these three stations than at

most or all other stations. Otherwise, *F* values for Error 1 effects were only greater than 1 for standing crop and richness, indicating that carry-over effects were limited for most benthic invertebrate community summary measures.

Relative Abundances Major Taxa

Table 5-26 provides rank correlations (r_s) between relative abundances of major taxa, and depth and distances. Large changes in correlations between years are of greatest importance. For n = 37 stations in each year, a difference in r_s of 0.46 would be significant at $p \leq 0.05$.

Table 5-26 Spearman Rank Correlations (r_s) Between Relative Abundances of Major Taxa and Depth and Distances from Drill Centres

X variable	Year	Relative abundance (*Y*)				
		Polychaeta	Bivalvia	Amphipoda	Tanaidacea	Echino-dermata
Depth	2000	0.081	0.055	–0.412*	0.112	0.012
	2004	**–0.643*** **	**0.712*** **	0.001	0.028	0.197
Distance from:						
Northern drill centre	2000	0.456**	–0.309	–0.248	–0.355*	0.052
	2004	–0.208	0.291	–0.301	–0.389*	0.341*
Southern drill centre	2000	–0.362*	0.230	–0.037	0.177	0.448**
	2004	0.006	–0.085	0.377*	0.102	–0.083
Central drill centre	2000	0.110	–0.175	–0.198	0.116	0.411*
	2004	–0.333*	0.300	0.015	0.028	0.156

Notes: - *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$ (in bold)
- n = 37 stations sampled in each year

As expected based on analysis of MDS1 scores, depth effects on polychaete and bivalve relative abundances were much greater in 2004 than in 2000 (Table 5-26). Correlations between abundances of these two taxa and distance from the Northern drill centre reversed between 2000 and 2004. There were also changes in correlations with distance from the Central and the Southern drill centres. These changes in distance correlations are presumably not drilling effects, since in both years, correlations with distance from the Northern and Southern drill centres were of opposite sign.

In 2000, but not in 2004, amphipod relative abundance decreased with increasing depth (Table 5-26; Figure 5-28). The regression lines based on untransformed original values in Figure 5-27 are provided as visual aids only, since the data and depth relationships were not suitable for parametric regression. Amphipod relative abundances were also uncorrelated with distance from the Southern drill centre in 2000, but positively correlated with distance from that drill centre in 2004 (Table 5-26; Figure 5-27). At distances of less than 2 km from the Southern drill centre, amphipod relative abundance in 2004 was lower than abundance in 2000 at the same stations. At distances greater than 2 km, abundances were similar between years.

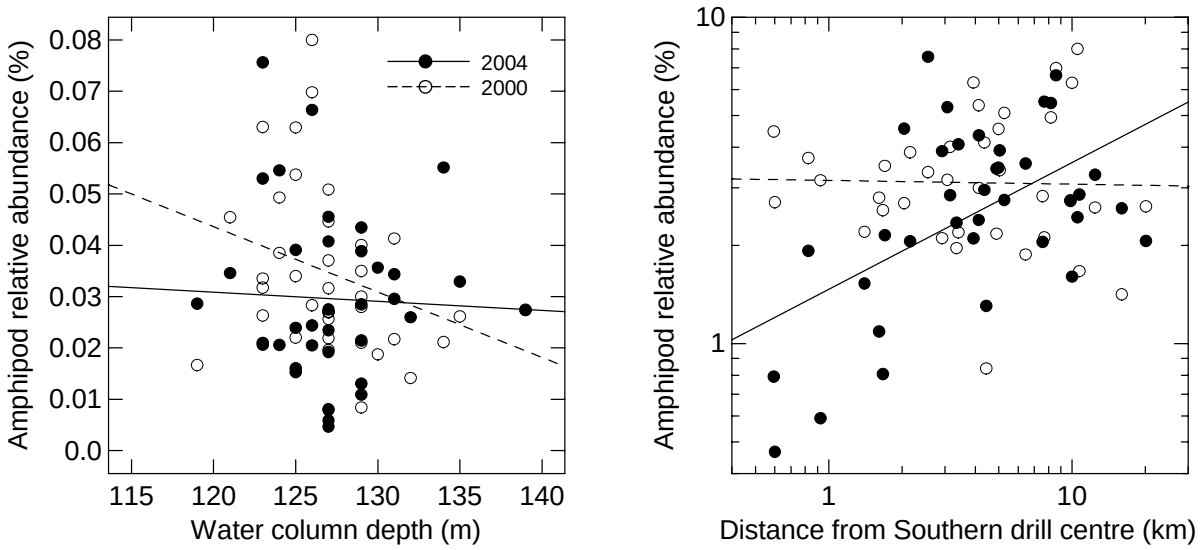


Figure 5-28 Amphipod Relative Abundance Versus Depth and Distance from the Southern Drill Centre (2000 and 2004)

Depth and distance correlations for Tanaidacea were similar between years (Table 5-26). The negative correlation with distance from the Northern drill centre observed for the full set of stations sampled in 2004 (Table 5-26) was also observed for the set of 37 stations sampled in both years, suggesting that it was a natural gradient.

Distance correlations for echinoderm relative abundance were generally the opposite of those for amphipod relative abundance (Table 5-26), because the two taxa accounted for most of the organisms that did not belong to the two dominant taxa (Polychaeta and Bivalvia). In 2000, echinoderm relative abundance was greater at more remote stations than near the location of the Southern drill centre (Figure 5-29; the lines are moving averages). In 2004, echinoderm relative abundance was more variable and was higher near the Southern drill centre than in 2000. This was probably not a stimulatory effect. Instead, with the relative abundance of amphipods depressed near the drill centre, the relative abundance of some other taxa must increase.

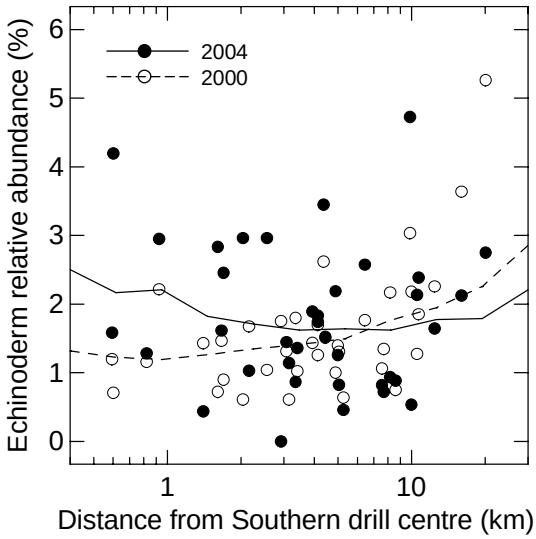


Figure 5-29 Echinoderm Relative Abundance Versus Distance from the Southern Drill Centre (2000 and 2004)

5.4.4 Integrated Assessment

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

Rank correlations between benthic invertebrate community variables and sediment particle size and organic carbon content were relatively weak and none were significant at $p \leq 0.001$ (Table 5-27). The expectation was that communities would be more diverse in sediments with higher gravel content than in uniformly sandy sediments (a habitat heterogeneity effect). This expectation was met to some extent, with all community variables except total abundance and relative abundance of polychaetes positively correlated with gravel content. However, any habitat heterogeneity effects were small relative to other effects (e.g., depth effects).

Table 5-27 Spearman Rank Correlations (r_s) Between Benthic Invertebrate Community Variables and Sediment Particle Size and Organic Carbon Content (2004)

Benthic Invertebrate Community Variable	Sediment Particle Size and Organic Carbon Content			
	% gravel	% sand	% fines	TOC
Summary Measures				
Total abundance	–0.190	0.158	0.116	–0.123
Standing crop	0.048	0.036	–0.198	–0.006
Richness	0.226	–0.262	0.163	0.038
Diversity	0.294*	–0.356**	0.235	0.165
Evenness	0.136	–0.186	0.107	0.230
MDS1	–0.181	0.264*	–0.289*	–0.206
MDS2	0.014	–0.063	0.321*	0.283*
Relative Abundances of Major Taxa				
Polychaeta	–0.269	0.306*	–0.224	–0.348**
Bivalvia	0.135	–0.209	0.328*	0.385**
Amphipoda	0.154	–0.130	–0.112	–0.139
Tanaidacea	0.078	–0.008	–0.090	0.070
Echinodermata	0.016	–0.009	–0.006	0.153

Notes: - $*p \leq 0.05$; $**p \leq 0.01$; $***p \leq 0.001$
- Richness, diversity, evenness and MDS scores are based families

Fines and TOC content positively affected richness, diversity and evenness, as well as bivalve:polychaete abundances (inverse of MDS1 scores) (Table 5-27). Most correlations were weak and a function of co-correlations with depth. The strong correlations between some benthic invertebrate community variables such as diversity, MDS1 scores, and relative abundances of polychaetes and bivalves (e.g., diversity) and depth were presumably not direct depth effects (e.g., due to variance in pressure) but indirect effects of some factor(s) correlated with depth. Fines content and several other physical and chemical variables were correlated with depth, and may have accounted for some of the apparent depth effects on community variables. However, correlations between those physical and chemical variables, either individually or collectively in multiple correlation analyses, and diversity, MDS1 scores, and relative abundances of polychaetes and bivalves were never as strong as correlations between the community variables and depth alone. Thus, the apparent depth effects were a function of some factor(s) unmeasured or poorly measured by the physical and chemical variables used in this and many other sediment quality studies.

5.4.4.2 Relationships Between Benthic Communities and Sediment Chemistry Variables

Rank correlations between benthic invertebrate community variables and sediment chemistry variables are provided in Table 5-28. The correlations with indicators of drilling activity ($>C_{10}$-C_{21} HCs, barium and possibly sulphur) can be considered measures of the strength (or weakness/absence) of exposure-response relationships between biology variables and contamination from drilling activity.

Table 5-28 Spearman Rank Correlations (r_s) Between Benthic Invertebrate Community Variables and Chemistry Variables (2004)

Benthic Invertebrate Community Variable	Sediment Chemistry Variable				
	>C_{10}-C_{21} HCs	**Barium**	**Sulphur**	**Metals PC1**	**Ammonia**
Summary Measures					
Total abundance	–0.171	–0.168	–0.098	–0.073	–0.036
Standing crop	–0.006	–0.017	0.060	0.245	0.148
Richness	–0.241	0.083	0.081	0.150	0.011
Diversity	–0.121	0.173	0.080	0.166	–0.007
Evenness	0.025	0.112	–0.025	0.077	0.014
MDS1	0.174	–0.283*	–0.219	–0.344*	0.043
MDS2	–0.039	–0.022	0.160	–0.047	–0.005
Relative Abundances of Major Taxa					
Polychaeta	0.106	–0.195	–0.258	–0.279	0.085
Bivalvia	0.012	0.340*	0.348**	0.382**	–0.159
Amphipoda	–0.432**	–0.429**	–0.375**	–0.363**	–0.005
Tanaidacea	–0.222	–0.151	0.067	–0.157	0.082
Echinodermata	0.109	0.087	0.130	0.153	0.076

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

Concentrations of >C_{10}-C_{21} HCs were not significantly correlated with any community variables except the relative abundance of amphipods (Table 5-28). The relative abundance of amphipods also decreased with increasing concentrations of barium and sulphur.

Diversity and the relative abundance of bivalves (correlated with MDS1 scores) and, to a lesser extent, richness and evenness, were positively correlated with concentrations of metals (including barium) and sulphur. Those correlations should not be considered positive or stimulatory effects of drilling on bivalves, but instead covariance of bivalve relative abundances and metals with some factor unrelated to drilling (e.g., depth or some unmeasured correlate with depth). In 2000, benthic community variables were also correlated with metal concentrations (Husky Energy 2001).

Ammonia was uncorrelated with community variables, despite the relatively wide range of ammonia concentrations.

5.5 Key Findings

5.5.1 Physical and Chemical Characteristics

Sediments collected from 56 stations in 2004 were predominantly (97.4%) sand. Fines (1.5%) and TOC content (0.105%) were low.

PAHs and BTEX were not detected in any sediment samples in 2004. >C_{10}-C_{21} HCs were detected at 45 of 56 stations at EQL = 0.025 mg/kg.

Aluminum, barium, chromium, iron, lead, manganese, strontium, thallium, uranium, vanadium and ammonia were detected at all 56 stations. Zinc was detected at 46 stations. Sulphur was detected at 55 stations.

TOC content was not significantly correlated with fines content. Metal, HC, sulphur and ammonia concentrations were weakly correlated with fines and TOC content, but these correlations were rarely significant. Barium and $>C_{10}$-C_{21} HC concentrations, used as indicators of drilling muds, were positively correlated. Barium concentrations were also positively correlated with concentrations of other metals which should be largely unaffected by drilling.

In 2004, concentrations of $>C_{10}$-C_{21} HCs and barium decreased significantly with distance from the Northern and Southern drill centres, where SBMs and WBMs were used. Contamination from the Southern drill centre was generally greater in magnitude and spatial extent than contamination from the Northern drill centre. In 2004, the median concentration of $>C_{10}$-C_{21} HCs was 22 mg/kg within 1 km of the Northern and Southern drill centres, and levels fell to approximately 1 mg/kg at distances of 5 km from these drill centres. Maximum concentration of $>C_{10}$-C_{21} HCs was 27.7 mg/kg and 275 mg/kg at the Northern and Southern drill centres, respectively. In contrast, all baseline (2000) concentrations of $>C_{10}$-C_{21} were below EQL (0.025 mg/kg). In 2004, low levels of $>C_{10}$-C_{21} HCs were also detected a three stations located more than 8 km from the drill centres (stations 11, 12, and 27; HC range: 0.42 to 0.66 mg/kg). However, these HCs did not have UCMs in the range of Puredrill IA-35 and PSC Maxxam reports that these HCs are probably non-petrogenic material.

Barium concentrations in 2004 generally reached background of less than 200 mg/kg within 2 km of the drill centres.

In 2004, both $>C_{10}$-C_{21} HCs and barium from the Southern drill centre were dispersed primarily to the southeast as opposed to the northwest, a directional effect not observed in 2000.

In 2004, fines content was elevated in the immediate vicinity of the Southern drill centre, increased with increasing depth and also increased, in general, from baseline (2000) values. In 2004, sulphur concentrations were also elevated in the immediate vicinity of the Southern drill centre and, to a lesser extent, in the immediate vicinity of the Northern drill centre. Fines content and sulphur concentrations in 2004 reached or approached background levels within 1 km of the drill centres.

In 2004, but not in 2000, concentrations of frequently detected metals other than barium decreased with distance from the Southern drill centre. Metal concentrations were higher in 2004 than in 2000 near that drill centre. However, concentrations of metals other than barium were lower in 2004 than in 2000 at intermediate and remote stations.

TOC and ammonia were largely unaffected by depth and distances from the Northern and Southern drill centres.

Carry-over effects, or persistent differences among stations over time (i.e., between 2000 and 2004) unrelated to depth or distance, were relatively small and rarely significant for sediment physical and chemical characteristics.

5.5.2 Toxicity

No sediment samples were toxic to either amphipods or bacteria in laboratory toxicity tests in 2000 and 2004.

5.5.3 Benthic Community Structure

Polychaetes accounted for approximately 75% of the invertebrates collected in 2000 and 2004 samples. Bivalves accounted for approximately 17% of the total. Amphipods, Tanaidacea and echinoderms were the only other major taxa accounting for more than 1% of total abundance in one or both years.

The primary patterns in community composition were related to the relative abundances of the two dominant taxa. When the relative abundance of polychaetes increased, the relative abundance of bivalves usually decreased, and vice versa. Diversity and, to a lesser extent, richness (number of families) were greater where polychaetes were less dominant. There was also a secondary difference between years. Cirratulidae (Polychaeta) were much more abundant at most stations in 2000 than in 2004, and Carditidae (Bivalvia) were collected at most stations in 2000 but at no stations in 2004.

In 2004, total abundance increased with increasing distance from the Northern and Southern drill centres. Distance effects did not extend beyond approximately 2 km from drill centres. However, the increases with distance from the Northern drill centre may have been natural. Similar increases were observed in 2000, although with fewer stations in the immediate vicinity of that drill centre. In contrast, in 2000, total abundance decreased with distance from the Southern drill centre (i.e., the reverse of the 2004 gradient).

In 2004, relative abundance of amphipods also increased with distance from the Southern. The distance effect did not extend beyond 2 km from the Southern drill centre and the distance gradient was not evident in 2000. The relative abundance of amphipods was also negatively correlated with concentrations of HCs.

Diversity and bivalve relative abundance increased with increasing depth in both sample years. Weaker relationships with depth were also observed for richness and evenness. None of these variables were affected by distance from the Northern or Southern drill centres. Standing crop was largely unaffected by depth and distance from the drill centres.

Carry-over effects, or persistent differences among stations over time (i.e., between 2000 and 2004) unrelated to depth or distance, were relatively weak for benthic invertebrate community variables and effects were significant only for bivalve:polychaete abundances (i.e., community composition).

5.5.4 Integrated Assessment

In 2004, all benthic invertebrate community variables except total abundance and the relative abundance of polychaetes were positively correlated with sediment gravel content, although the correlations were weak and rarely significant. Richness, diversity, evenness and the relative abundance of bivalves were also positively correlated with fines and TOC content. Those correlations were relatively weak and probably reflected co-correlations of the variables with depth.

In 2004, the relative abundance of amphipods was significantly negatively correlated with concentrations of the two drilling indicators, $>C_{10}$-C_{21} HCs and barium. The relative abundance of amphipods was also negatively correlated with sulphur. Other benthic community variables were not significantly correlated with $>C_{10}$-C_{21} HC concentrations. Richness, evenness and the relative abundance of bivalves were positively correlated with barium and sulphur concentrations but these variables were also correlated with concentrations of metals other than barium.

Ammonia concentrations were uncorrelated with benthic invertebrate community variables.

6.0 Commercial Fish Component

6.1 Field Collection

The *CCG Wilfred Templeman*, its crew and DFO Science personnel were chartered for the 2004 commercial fish survey of American plaice ("plaice") and snow crab ("crab") between July 10 and July 18, 2004. Collection dates for the baseline program, and tests performed on collected specimens, are shown in Table 6-1.

Table 6-1 Field Trips Dates

Trip	Collections/Tests	Date
Baseline Program	Study Area Crab for Body Burden Analysis; Study and Reference Area American plaice for body burden and taste analysis; Study Area plaice for health analysis.	July 4 to July 10, 2000
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

Notes:
- Since the location of Reference Areas sampled in 2004 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 and 2002 samples are presented in Husky Energy (2001; 2003). Sampling for the 2004 program was conducted under a Department of Fisheries and Oceans Stock Assessment license. A total of 85 plaice and 63 crab were collected in the White Rose Study Area in 2004. A total of 136 plaice and 85 crab were collected across four Reference Areas. Location of transects are provided in Figure 6-1 and Appendix C-1. 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.

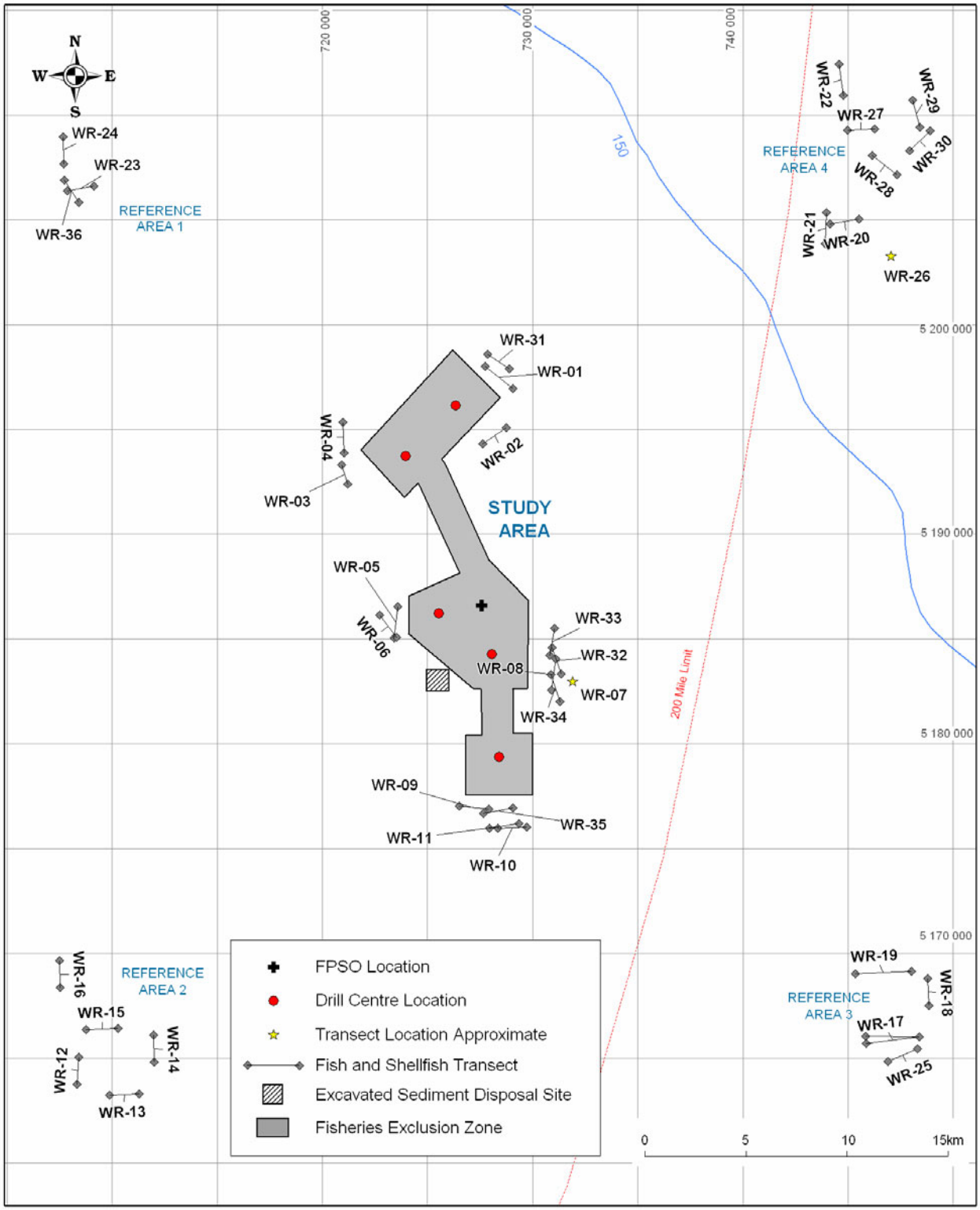


Figure 6-1 Plaice and Crab Transects

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 smaller female crab.

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 two blood smears were prepared for each fish 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 Mixed Function Oxygenase (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 14 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 focussed on individual fish rather than composite or randomly assigned samples (Table 6-2).

Table 6-2 Plaice Selected for Body Burden, Taste and Health Analyses (2004)

Transect Number	Group	Total No. Fish	Body Burden Composites (Bottom Fillet, or Liver)	Taste (Number of Fish, Top Fillet)	Health (Number of Fish)
WR-01	Study	7	Composite 1 (7 fish)	2	7
WR-02	Study	8	Composite 2 (8 fish)	2	8
WR-03	Study	8	Composite 3 (8fish)	2	8
WR-04	Study	7	Composite 4 (7 fish)	2	7
WR-05	Study	6	Composite 5 (5 fish)	2	5
WR-06	Study	7	Composite 6 (7 fish)	2	7
WR-07	Study	5	Composite 7 (5 fish)	2	5
WR-08	Study	9	Composite 8 (6 fish)	2	6
WR-09	Study	3	Composite 9 (8 fish)	2	3
WR-35	Study	19			5
WR-10	Study	0		0	0
WR-11	Study	6	Composite 10 (6 fish)	2	0
Total	**Study**	**85**	**10**	**20**	**61**
WR-12	Reference 2	10	Composite 11 (15 fish)	3	9
WR-13	Reference 2	2			2
WR-14	Reference 2	5			4
WR-15	Reference 2	9	Composite 12 (9 fish)	2	9
WR-16	Reference 2	9	Composite 13 (9 fish)	2	9
WR-17	Reference 3	7	Composite 14 (7 fish)	2	7
WR-18	Reference 3	9	Composite 15 (9 fish)	2	9
WR-19	Reference 3	16	Composite 16 (13 fish)	2	13
WR-20	Reference 4	12	Composite 17 (11 fish)	2	11
WR-21	Reference 4	9	Composite 18 (9 fish)	2	9
WR-22	Reference 4	11	Composite 19 (11 fish)	2	11
WR-23	Reference 1	9	Composite 20 (9 fish)	2	9
WR-24	Reference 1	12	Composite 21 (12 fish)	2	12
WR-36	Reference 1	16	Composite 22 (5 fish)	2	5
Total	**Reference**	**136**	**12**	**25**	**119**

Notes:
- Sixty-one fish were selected for health analyses in the Study Area and 119 were selected from the Reference Areas
- For the most part, those fish excluded from health analysis were also excluded from body burden and taste analysis
- However, trawl WR-11 which was not sampled for health, was required for body burden to achieve 10 composites for the Study Area
- Trawl WR-11 was also used in taste analysis

Crab from 16 trawls in the Study Area and 20 trawls in the Reference Areas were used for body burden and taste analysis. Tissue from right legs were composited to generate 10 individual body burden samples for the Study Area and two to three individual samples for each of the four

Reference Areas (Table 6-3). Left leg tissue from each trawl used in body burden analysis 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 of Crab, Left Legs)
WR-01	Study	3	Composite 1 (4 crab)	3
WR-02	Study	1		
WR-03	Study	6	Composite 2 (6 crab)	3
WR-04	Study	5	Composite 3 (5 crab)	3
WR-05	Study	8	Composite 4 (8 crab)	3
WR-06	Study	7	Composite 5 (7 crab)	3
WR-07	Study	0		0
WR-08	Study	3	Composite 6 (7crab)	3
WR-34	Study	4		
WR-09	Study	3	Composite 7 (5 crab)	3
WR-10	Study	2		
WR-11	Study	5	Composite 8 (5 crab)	3
WR-31	Study	8	Composite 9 (8 crab)	3
WR-32	Study	2	Composite 10 (8 crab)	3
WR-33	Study	6		
WR-35	Study	0		0
Total	**Study**	**63**	**10**	**30**
WR-12	Reference 2	3	Composite 11 (5 crab)	3
WR-13	Reference 2	2		
WR-14	Reference 2	2	Composite 12 (5 crab)	3
WR-15	Reference 2	3		
WR-16	Reference 2	4	Composite 13 (4 crab)	3
WR-17	Reference 3	2	Composite 14 (7 crab)	3
WR-18	Reference 3	2		
WR-19	Reference 3	3		
WR-20	Reference 4	10	Composite 15 (10 crab)	3
WR-21	Reference 4	10	Composite 16 (10 crab)	3
WR-22	Reference 4	2	Composite 17 (12 crab)	3
WR-26	Reference 4	1		
WR-27	Reference 4	1		
WR-30	Reference 4	8		
WR-23	Reference 1	7	Composite 18 (7 crab)	3
WR-24	Reference 1	4	Composite 19 (4 crab)	3
WR-25	Reference 3	12	Composite 20 (12 crab)	3
WR-28	Reference 4	0		0
WR-29	Reference 4	0		0
WR-36	Reference 1	9	Composite 21 (9 crab)	3
Total	**Reference**	**85**	**11**	**33**

Note: - Numbers approximate because crab legs were often broken off carapace

6.2.2 Body Burden

Samples were delivered frozen to PSC 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 2004)

Variables	Method	2000 EQL	2002 EQL	2004 EQL	Units
Hydrocarbons					
$>C_{10}$-C_{21}	GC/FID	15	15	15	mg/kg
$>C_{21}$-C_{32}	GC/FID	15	15	15	mg/kg
$>C_{10}$-C_{32}	Calculated	30	30	30	mg/kg
PAHs					
1-Chloronaphthalene	GC/MS	NA	NA	0.05	mg/kg
2-Chloronaphthalene	GC/MS	NA	NA	0.05	mg/kg
1-Methylnaphthalene	GC/MS	0.05	0.05	0.05	mg/kg
2-Methylnaphthalene	GC/MS	0.05	0.05	0.05	mg/kg
Acenaphthene	GC/MS	0.05	0.05	0.05	mg/kg
Acenaphthylene	GC/MS	0.05	0.05	0.05	mg/kg
Anthracene	GC/MS	0.05	0.05	0.05	mg/kg
Benz[a]anthracene	GC/MS	0.05	0.05	0.05	mg/kg
Benzo[a]pyrene	GC/MS	0.05	0.05	0.05	mg/kg
Benzo[b]fluoranthene	GC/MS	0.05	0.05	0.05	mg/kg
Benzo[ghi]perylene	GC/MS	0.05	0.05	0.05	mg/kg
Benzo[k]fluoranthene	GC/MS	0.05	0.05	0.05	mg/kg
Chrysene	GC/MS	0.05	0.05	0.05	mg/kg
Dibenz[a,h]anthracene	GC/MS	0.05	0.05	0.05	mg/kg
Fluoranthene	GC/MS	0.05	0.05	0.05	mg/kg
Fluorene	GC/MS	0.05	0.05	0.05	mg/kg
Indeno[1,2,3-cd]pyrene	GC/MS	0.05	0.05	0.05	mg/kg
Naphthalene	GC/MS	0.05	0.05	0.05	mg/kg
Perylene	GC/MS	0.05	0.05	0.05	mg/kg
Phenanthrene	GC/MS	0.05	0.05	0.05	mg/kg
Pyrene	GC/MS	0.05	0.05	0.05	mg/kg
Metals					
Aluminum	ICP-MS	2.5	2.5	2.5	mg/kg
Antimony	ICP-MS	0.5	0.5	0.5	mg/kg
Arsenic	ICP-MS	0.5	0.5	0.5	mg/kg
Barium	ICP-MS	1.5	1.5	1.5	mg/kg
Beryllium	ICP-MS	1.5	1.5	0.5	mg/kg
Boron	ICP-MS	1.5	1.5	1.5	mg/kg
Cadmium	GFAAS	0.08	0.05	0.05	mg/kg
Chromium	ICP-MS	0.5	0.5	0.5	mg/kg
Cobalt	ICP-MS	0.2	0.2	0.2	mg/kg
Copper	ICP-MS	0.5	0.5	0.5	mg/kg
Iron	ICP-MS	5	5	15	mg/kg
Lead	ICP-MS	0.18	0.18	0.18	mg/kg
Lithium	ICP-MS	0.5	0.5	0.5	mg/kg
Manganese	ICP-MS	0.5	0.5	0.5	mg/kg
Mercury	CVAA	0.01	0.01	0.01	mg/kg
Molybdenum	ICP-MS	0.5	0.5	0.5	mg/kg
Nickel	ICP-MS	0.5	0.5	0.5	mg/kg

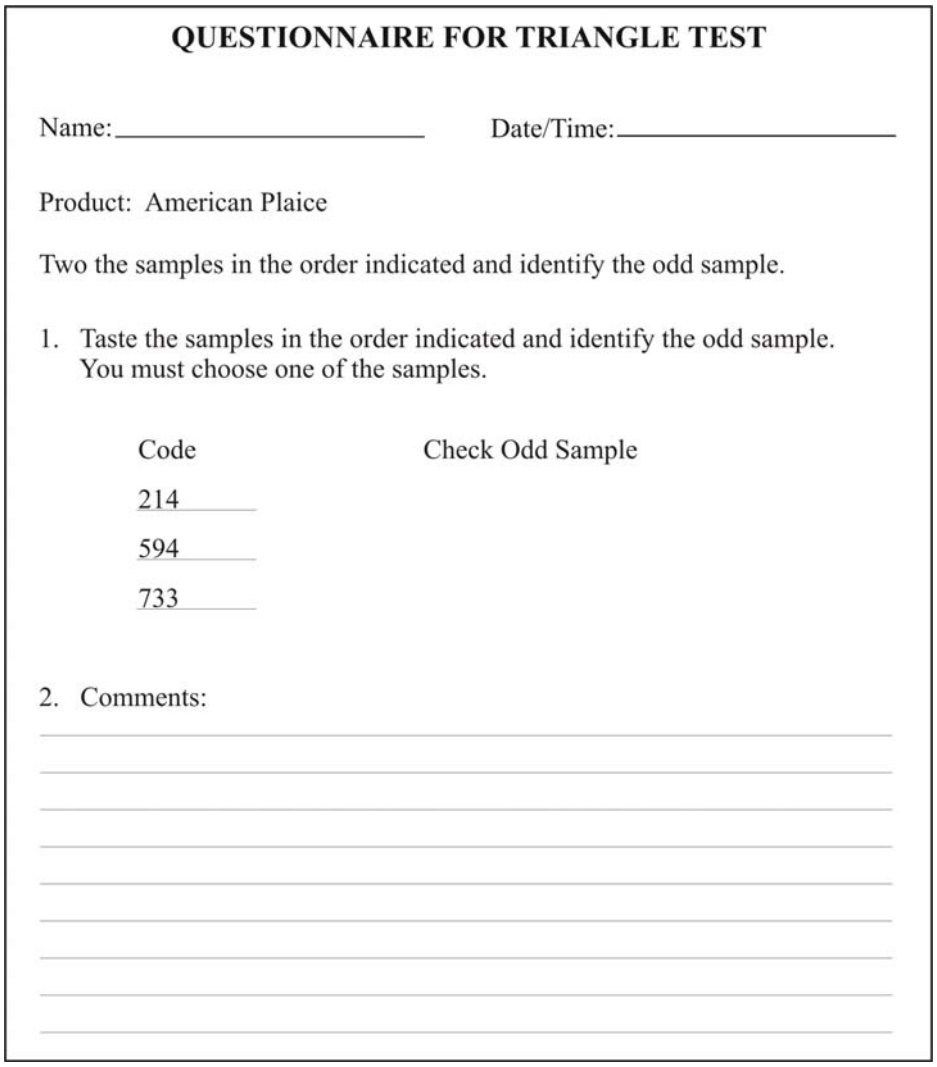
Variables	Method	2000 EQL	2002 EQL	2004 EQL	Units
Selenium	ICP-MS	0.5	0.5	0.5	mg/kg
Silver	ICP-MS	0.12	0.12	0.12	mg/kg
Strontium	ICP-MS	1.5	1.5	1.5	mg/kg
Thallium	ICP-MS	0.02	0.02	0.02	mg/kg
Tin	ICP-MS	0.5	0.5	0.5	mg/kg
Uranium	ICP-MS	0.02	0.02	0.02	mg/kg
Vanadium	ICP-MS	0.5	0.5	0.5	mg/kg
Zinc	ICP-MS	0.5	0.5	0.5	mg/kg
Other					
Percent Lipids	PEI FTC	0.1	NA	NA	%
Crude Fat	AOAC922.06	NA	0.5	0.5	%
Moisture	Grav.	0.1	0.1	0.1	%

Notes:
- The EQL is the lowest concentration that can be reliably achieved 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. Frozen samples were thawed for 24 hours at 2°C and allocated to either the triangle taste test or the hedonic scaling test. 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. Samples were then rinsed, enclosed in individual aluminum foil packets (shiny side in), labeled with a predetermined random three-digit code, cooked in a convection oven at 175°C for 15 minutes and then served at 35°C.

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. Each panelist was provided with a cup of room-temperature water for rinsing and a cup for expectorate. Panelists were instructed not to communicate with each other while in the panel room (Figure 6-4) and to leave immediately upon completion of the taste tests.



QUESTIONNAIRE FOR TRIANGLE TEST

Name:_____ Date/Time:_____

Product: American Plaice

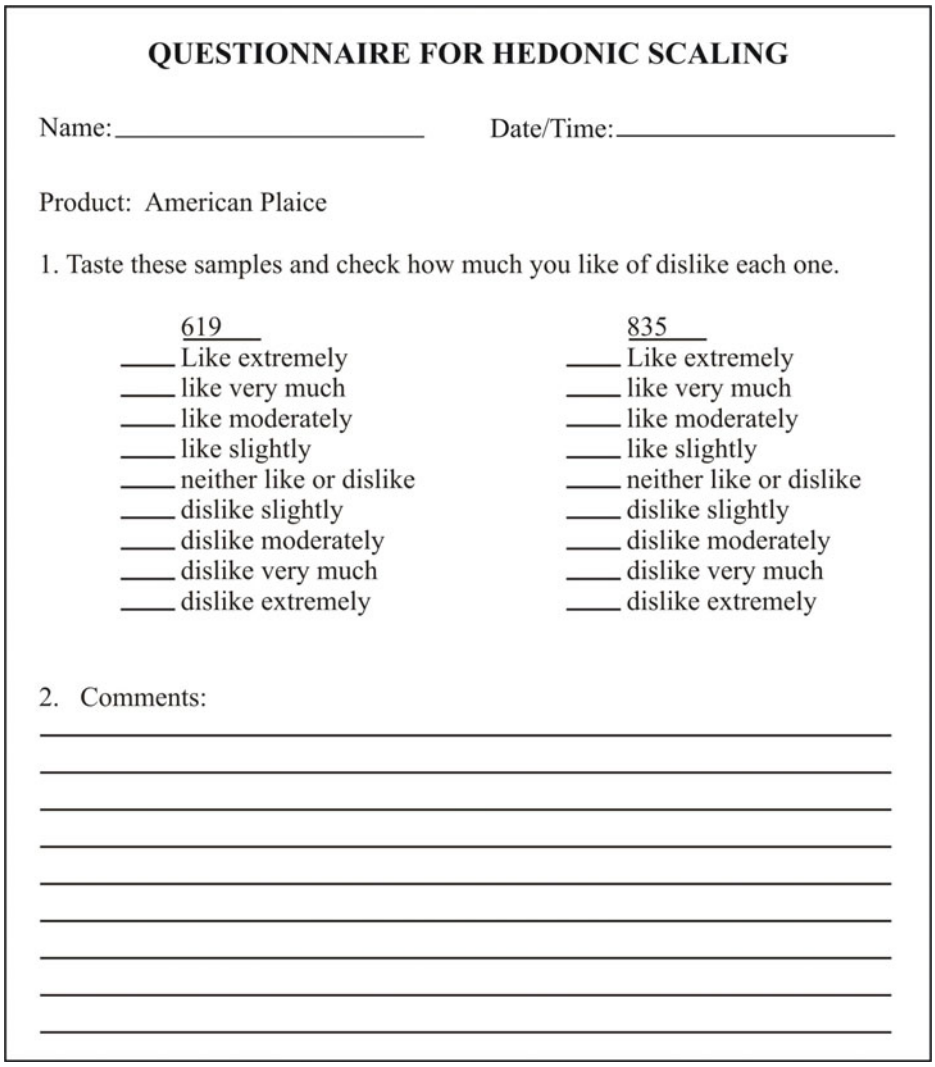
Two the samples in the order indicated and identify the odd sample.

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.

619	835
____ Like extremely	____ Like extremely
____ like very much	____ like very much
____ like moderately	____ like moderately
____ like slightly	____ like slightly
____ neither like or dislike	____ neither like or dislike
____ dislike slightly	____ dislike slightly
____ dislike moderately	____ dislike moderately
____ dislike very much	____ dislike very much
____ dislike extremely	____ dislike extremely

2. Comments:

Figure 6-3 Questionnaire for Sensory Evaluation by Hedonic Scaling

Figure 6-4 Panel Room for Taste Tests

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 Assay

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°C and homogenized in four volumes of 50 mM Tris buffer, pH 7.5, (1 g liver to 4 ml buffer) using at least 10 passes of a glass Ten Broek hand homogenizer. Homogenates were centrifuged at 9,000 g for 15 minutes at 4°C and the post-mitochondrial supernatant (S9 fraction) was frozen in triplicate at -65°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 µM ethoxyresorufin (Sigma) dissolved in dimethyl sulphoxide, 0.15 mM NADPH and 20 µl of S9 protein (diluted 5 times). After a 15-minute incubation at 27°C, the reaction was stopped with 2 ml of methanol (HPLC grade) and samples were centrifuged (3,600 g for 5 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 at the beginning of 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 the 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 previous descriptions (Ellis 1976). Because blood cells do not disperse randomly on a slide when a smear is made, the standard procedure, Exaggerated Battlement Method (EBM), was performed to ensure that cells in one particular area (i.e., the middle or the edges of the slide) were not missed (Lynch et al. 1969).

6.2.4.3 Tissue 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 the 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
2. Megalocytic hepatosis
3. Eosinophilic foci
4. Basophilic foci
5. Clear cell foci
6. Hepatocellular carcinoma
7. Cholangioma
8. Cholangiofibrosis
9. Macrophage aggregates
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 lesion 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) *tip hyperplasia* - thickening of the epithelium at the very tip of lamellae giving the appearance of a club; and b) *telangiectasis* - 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 White Rose EEM program used a multiple-reference design, with four Reference Areas and a single Study Area. Two comparisons (contrasts) were of interest:

- Study versus Reference (SR contrast); and
- Among References.

Table 6-6 provides the basic nested Analysis of Variance (ANOVA) used to test these contrasts. The Study versus Reference (SR) contrast is tested against the variance among Reference Areas or MS(A{R}). The four Reference Areas, not composites or individual crab or plaice within areas, are the appropriate replicates for testing the SR contrast. The test is equivalent to a *t* test or ANOVA comparing the Study Area mean to the sample of four Reference Area means (Sokal and Rohlf 1981, p. 231), although results (*p* values) for the two approaches will be identical when sample sizes are the same in all areas.

Table 6-6 Nested ANOVA Model for Analysis of Multiple-Reference Design, with Four Reference Areas

Source/Term	df	Mean Square (MS)	*F*
Among Areas			
Study versus Reference (SR)	1	MS(SR)	MS(SR)/MS(A{R})
Areas within Reference (A{R})	3	MS(A{R})	MS(A{R})/MSE
Within Areas			
Among Composites	*N*–5	MSE	

Note: - *N* = total number of composites; *N* = 21 for crab ; *N* = 22 for plaice

The Among Reference Area contrast is tested against the variance among replicates within areas (MSE). The test of the Among Reference Area contrast is equivalent to a comparison of the four Reference Areas in a one-way ANOVA, except that variance within the Study Area is also incorporated into the MSE.

With four replicate Reference Areas, the test of the SR contrast has limited power. If the variance among Reference Areas is "small" (i.e., MS(A{R}) $\approx$ MSE), power can be increased by testing the SR contrast against the MSE, based on 21 or 22 composites. Winer (1971) recommended testing against the MSE in nested ANOVA when $p > 0.20$ for the Among Reference Area contrast. His recommendation was adopted in this report for interpretation. However, *p* for tests against both MS(A{R}) and MSE are provided, since other *p* values (i.e., 0.05 to >> 0.50) could be used to define MS(A{R}) as "small" (Sokal and Rohlf 1981).

Data and residuals for parametric analyses (ANOVA and Principal Component Analyses) were graphically screened to identify departures from normality and homogeneity of variances, and outliers. These departures were addressed either by using transformations or non-parametric tests, or by identifying cases and providing warnings when test results (typically *p* values) might not be robust or precise regardless of the test or transformation used. The standard $p \leq 0.05$ was used to define statistical significance, but that is an arbitrary choice. Realistically, $0.01 < p < 0.10$ should usually be regarded as ambiguous, because the precise values of *p* will be partly to largely dependent on the tests, error terms and transformations used.

6.3.1 Biological Characteristics of Crab and Plaice

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

6.3.1.1 Crab

Biological Characteristics of crab analyzed were carapace width, claw (chela) height, and frequency of recent (current year or 2004) moults based on measures of 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 (2+ years since moult). Values other than 1 to 4 and 6 were not observed.

The first step in analysis of crab Biological Characteristics 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 crab within composites), was used for the analysis. The variance among crab within composites is the error term for testing variance among composites within areas. Recent moults were scored as 0 and non-recent moults as 1 for the analysis. Results should be regarded as approximate, since only two values were possible. Equivalent nested tests based on frequencies or counts (e.g., χ^2 or log-likelihood {*G*} tests) are not known. Analyses were repeated using the Reference Areas only (again, a three-level nested ANOVA) and the Study Area only (one-way ANOVA comparing composites).

The above analyses indicated that there was significant added variance among composites within areas for all three variables. Therefore, mean carapace width and chela height, and the frequency of recent moults, were calculated for each composite, and the composite values analyzed in the nested ANOVA in Table 6-6. Frequencies of recent moults were rank-transformed.

Spearman rank correlations (r_S) were also calculated among the three biological variables, based on individual and composite values. Correlations were calculated over all areas pooled, pooled Reference Areas and within the Study Area.

6.3.1.2 Plaice

In this section, analyses of plaice Biological Characteristics were restricted to gutted weight (i.e., size). Males, immature females and mature spent females were pooled for all analyses, since they were also pooled within the composites used for body burden analyses. Again, the primary objective was to determine if there were size differences that might affect analyses of body burdens. Appendix C-4 provides more extensive analyses of a larger suite of biological variables (length, age, liver weight, gonad weight, etc.).

All analyses in this section 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. The distributions of individual weights were also truncated at the left (low) end since fish smaller than 250 mm in length were released. Composite mean weights were compared among areas using the nested model in Table 6-6.

6.3.2 Body Burden

6.3.2.1 Crab

Summary statistics for body burdens from crab collected in the Study and Reference Areas in 2004 were generated and body burdens from the Study Area in 2004 were qualitatively compared to Study Area values in 2000.

Additional analyses of 2004 body burdens were conducted on moisture, fat content and dry weight concentrations of the eight metals detected in all or most composites (arsenic, boron, copper, mercury, selenium, silver, strontium, zinc). Values less than EQL were set at ½ EQL.

A summary measure of metal concentrations was derived using PCA. PCA identifies the major axis of covariance (= Principal Components or PC1) among the original variables (concentrations of the eight metals), which is also the major axis of variance among samples (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 count 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 variable values. The scores are usually scaled so that the mean is 0 and the variance and standard deviation (SD) are 1. These scores can be used as summary variable values for subsequent analyses. In this study, metal concentrations were $\log_{10}$ transformed prior to conducting the PCA. Only PC1 scores were retained for further analyses, since PC2 and lesser PCs accounted for a limited amount of variance.

Fat and moisture content, Metals PC1, and concentrations of the eight individual metals were analyzed in the nested ANOVA in Table 6-6. Rank correlations were also calculated among body burden variables and between body burden variables and the three biological variables (carapace width, claw height, % recent moults).

6.3.2.2 Plaice

Liver
Summary statistics for liver body burdens from plaice collected in the Study and Reference Areas in 2004 were generated, and body burdens from the Study Area in 2004 were qualitatively compared to Study Area values in 2000.

Additional analyses on 2004 liver body burden variables were conducted on moisture, fat content, concentrations of eight metals detected in all composites (arsenic, cadmium, copper, iron, manganese, mercury, selenium and zinc) and concentrations of compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} range.

PCA was used on log-transformed metal concentrations to derive two summary measures (Metals PC1 and PC2). Moisture and fat content, $>C_{10}$-C_{21} and C_{21}-C_{32} concentrations, Metals PC1 and PC2 scores, and concentrations of the eight individual metals were compared among areas using the nested ANOVA in Table 6-6. Spearman rank correlations (r_S) were also calculated among moisture and fat content and Metals PC1, and between those variables and composite mean weights.

Fillet
Summary statistics for fillet body burdens from plaice collected in the Study and Reference Areas in 2004 were generated, and body burdens from the Study Area in 2004 were qualitatively compared to Study Area values in 2000.

Additional analyses on 2004 fillet body burdens were conducted on fillet moisture and fat content, and concentrations of arsenic, mercury and zinc, the only frequently detected metals. Analyses were the same as for liver variables, except that PCA was unnecessary with only three metals.

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-3 (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 single Study Area was used in analyses and two comparisons, Study versus Reference and Among References, 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-4 (Annex B).

6.4 Results

6.4.1 Biological Characteristics of Crab and Plaice

6.4.1.1 Crab

Summary statistics for carapace width and claw height based on individual crab are provided for each area in Table 6-7. Overall, 85 Reference and 63 Study Area crab were used for body burden analyses, although claw height was not measured on a few crab with damaged or missing chelae. Medians and means of the Reference Area means are also provided in Table 6-7 for comparison to Study Area means. Crab were largest in Reference Area 4. Study Area crab were larger than crab from Reference Area 1 and 2, and similar in size to crab from Reference Area 3. The SDs and Coefficient of Variations (CVs) for the two size variables in Reference Area 4 were approximately half the SDs and CVs in other areas. Restricting samples and analyses to crab larger than 40 cm carapace width did not truncate size distribution, as most crab were much larger than 40 cm width (Table 6-7).

Table 6-7 Summary Statistics for Individual Crab Carapace Width and Chela (Claw) Height

Variable	Area	*n*	Min	Max	Median	Mean	SD	CV (%)
Carapace width (mm)	Reference 1	20	41	125	101	92	26	28
	Reference 2	14	48	123	90	87	23	26
	Reference 3	19	51	126	102	100	24	24
	Reference 4	32	85	132	115	112	11	10
	Reference means				96	98		
	Study	63	46	160	108	103	22	22
Claw height (mm)	Reference 1	20	6.5	30.5	22.3	20.7	7.6	37
	Reference 2	13	7.0	31.0	17.0	17.4	6.8	39
	Reference 3	15	10.5	34.0	25.1	24.2	7.6	31
	Reference 4	31	18.2	34.0	27.9	26.6	3.8	14
	Reference means				22.5	22.3		
	Study	61	6.5	35.5	26.2	23.8	7.3	31

Note: - CV = Coefficient of Variation (SD as % of mean)

Frequencies of shell condition values are provided in Table 6-8. Index values of 1 to 4 and 6 were the only values observed, and values of 1 and 4 were rare. Based on these values, Reference Area 4 crab were unusual, in that only one crab had apparently moulted in 2004 (recent moult). In contrast, 36 to 86% of crab in other areas were recent moults. Most (20) of the Reference Area crab had not moulted in the past year (2003; index value = 6), although that was also true for most of the Study Area non-recent moults.

Table 6-8 Frequencies of Crab Shell Condition Index Values

Moult year	Index value	Area						Total
		Ref 1	Ref 2	Ref 3	Ref 4	All refs	Study	
Recent (0)	1	0	1	0	0	1	5	8
	2	10	11	8	1	30	20	82
Total (No.)		10	12	8	1	31	25	56
(%)		50	86	42	3	36	40	38
Not recent (–1+)								
Last year (–1)	6	4	0	8	11	23	10	62
Previous (–2+)	3	6	2	3	20	31	26	91
	4	0	0	0	0	0	2	6
Total (No.)		10	2	11	31	54	38	92
(%)		50	14	58	97	64	60	62
Grand total (No.)		20	14	19	32	85	63	148
(%)		100	100	100	100	100	100	100

Notes: - Moult years: 0 = 2004; –1 = 2003; –2+ = 2002 or earlier
- Values are numbers of crab unless otherwise indicated

The three biological variables differed significantly among composites within areas (Table 6-9). Except for % recent moult, the significant differences among composites occurred within the Reference Areas but not within the Study Area. Those results were surprising, since the Reference Area composites and trawls were collected in small areas. The differences within Reference Areas occurred mostly in Reference Areas 1 to 3. Within Reference Area 4, there was limited variance of carapace or claw size at any level (Table 6-7), and almost no variance in % recent moult (i.e., only one crab was a recent moult; Table 6-8). Consequently, variances within composites were unequal

for the comparison of all areas, and the four Reference Areas, and the *p* in Table 6-9 approximate (although most *p* were either << 0.01 or >> 0.10).

Table 6-9 Results (*p*) for Comparisons of Crab Biological Characteristics Among Composites Within Areas

Variable	All Areas	Reference Areas	Study Area
Carapace width	0.006	< 0.001	0.455
Claw height	0.028	0.003	0.448
% recent moult	< 0.001	0.004	< 0.001

Summary statistics for composite means are provided in Table 6-10. Mean carapace and claw sizes were similar to those based on individual crab (Table 6-7). Minima were higher, maxima lower, and SDs and CVs lower because composite means vary less than individual values. Except for the Study Area, medians are not robust, since they were based on only two or three composites. CVs are not provided for % recent moult, because values and the mean could be expressed as % recent moult or as % non-recent moult (100–% recent moult; SD remain the same). For all three variables, SDs and CVs were much lower for Reference Area 4 than for other areas. SDs and CVs for carapace and claw size for the Study Area were lower than for Reference Areas 1 to 3, because variance in the Study Area was mostly within rather than among composites (Table 6-9). These differences in variance among composites within areas affected comparisons of composite means among areas (see below).

Table 6-10 Summary Statistics for Biological Characteristic of Crab, Based on Composite Means

Variable	Area	*n*	Min	Max	Median	Mean	SD	CV (%)
Carapace width (mm)	Reference 1	3	75	108	103	95	18	18
	Reference 2	3	72	97	95	88	14	15
	Reference 3	2	84	110	97	97	18	19
	Reference 4	3	109	114	113	112	3	2
	Reference means				96	98		
	Study	10	90	115	100	103	9	
Claw height (mm)	Reference 1	3	16.0	25.3	23.3	21.5	4.8	23
	Reference 2	3	14.3	20.7	17.2	17.4	3.2	18
	Reference 3	2	18.4	27.2	22.8	22.8	6.2	27
	Reference 4	3	25.2	27.4	27.2	26.6	1.2	4
	Reference means				22.1	22.1		
	Study	10	18.8	28.1	23.0	23.6	2.9	12
% recent moult	Reference 1	3	14	89	25	43	40	
	Reference 2	3	80	100	80	87	12	
	Reference 3	2	33	57	45	45	17	
	Reference 4	3	0	8	0	3	5	
	Reference means				44	44		
	Study	10	0	88	27	39	41	

Note: - CV = Coefficient of Variation (SD as % of mean)

None of the three biological variables differed significantly between the Reference and Study Areas (*p* for SR contrast in Table 6-11). Instead, the largest differences occurred among the Reference Areas, with Reference Area 4 crab larger and further from last moult than crab from other areas (Table 6-10). Those general conclusions should be regarded as robust, although the *p* values in Table 6-11 are approximate because variances among composites were not equal among areas (see above; Table 6-10). The Study versus Reference (SR) contrast was never significant regardless of whether it was tested against the variance among Reference Areas or the variance among composites within areas (all $p >> 0.10$).

Table 6-11 Results (*p*) of Nested ANOVA Comparing Biological Characteristics of Crab Among Areas

Variable	Contrast		
	Among References	Study versus Reference (SR)	
		Error=	
		Among References	Among composites
Carapace width	0.113	0.576	0.354
Claw height	**0.033**	0.640	0.333
% recent moult[1]	0.097	0.766	0.613

Notes: - Table 6-6 and Section 6.3 provide details on the nested ANOVA and contrasts
- $p \leq 0.05$ (in bold)
- [1] rank-transformed

As expected, individual values and composite means for the two size measures (carapace width and claw height) were strongly positively correlated over all areas, within Reference Areas, and within the Study Area (Table 6-12). The two size measures were negatively correlated with % recent moult, indicating that smaller crab were more likely to have moulted in 2004.

Table 6-12 Spearman Rank Correlations (r_s) Among Crab Biological Variables

Values	Areas	Carapace width-claw height		Carapace width-% recent moult		Claw height-% recent moult	
		n	r_s	*n*	r_s	*n*	r_s
Individual crab	All	140	**0.944****	148	**–0.346****	140	**–0.390****
	Reference	79	**0.921****	85	**–0.390****	79	**–0.457****
	Study	61	**0.962****	63	**–0.276***	61	**–0.315***
Composite means	All	21	**0.942****	21	**–0.677****	21	**–0.713****
	Reference	11	**0.964****	11	**–0.781****	11	**–0.817****
	Study	10	**0.903****	10	–0.500	10	–0.538

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

6.4.1.2 Plaice

Females accounted for 80 to 90% of the catch in each area. Most composites were composed of two size classes: small immature fish usually less than 300 g and larger mature fish that could exceed 1,000 g (Figure 6-5). Because of this bimodal size distributions, all analyses were based on composite mean weights.

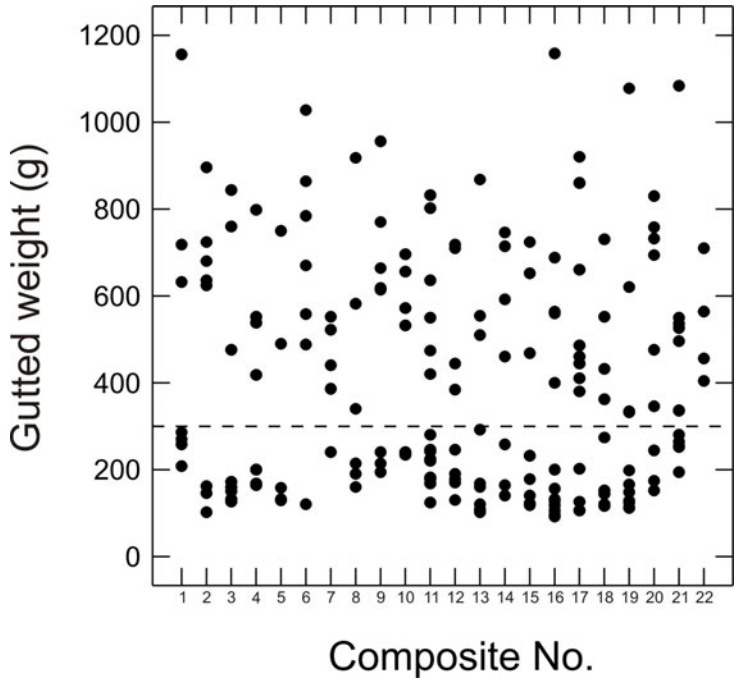


Figure 6-5 Distribution of Plaice Gutted Weights Within Composites
Note: some points may represent more than one fish

Summary statistics of composite mean weights are provided in Table 6-13. Study Area plaice were larger than those from all Reference Areas except Reference Area 1.

Table 6-13 Summary Statistics for Plaice Gutted Weight, Based on Composite Means

Area	*n*	Min	Max	Median	Mean	SD	CV (%)
Reference Area 1	3	426	540	490	485	57	12
Reference Area 2	3	320	372	352	348	26	7
Reference Area 3	3	318	439	338	365	65	18
Reference Area 4	3	305	459	320	361	85	24
Reference means				345	390		
Study	10	332	645	458	459	94	20

Notes: - Units for weight are g
- CV = Coefficient of Variation (SD as % of mean)

Differences in composite mean weight between the Study and Reference Areas were not significant, regardless of whether they were tested against the variance among Reference Areas (p = 0.243) or the variance among composites within areas (p = 0.063).

Similarly, there were no significant differences for most of the other biological variables (length, age, etc.) tested in conjunction with fish health analyses (Appendix C-4). Gutted weight to length, however, was greater in the Study Area for immature females when variance among areas was used as the error term (Appendix C-4, Table 9).

6.4.2 Body Burden

6.4.2.1 Crab

Summary statistics for concentrations of detected substances in crab claw composites in 2004 are provided in Table 6-14. Summary statistics for detected substances in the Study Area in 2000, and comparison to 2004 data, are provided in Table 6-15.

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

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV %
Arsenic	Reference 1	3	0	6.90	7.80	7.50	7.40	0.46	6
	Reference 2	3	0	6.80	10.00	9.60	8.80	1.74	20
	Reference 3	2	0	8.50	8.60	8.55	8.55	0.07	1
	Reference 4	3	0	11.00	13.00	11.00	11.67	1.15	10
	Reference Means					9.16	9.10		
	Study	10	0	4.80	12.00	8.55	8.71	2.44	28
Boron	Reference 1	3	0	1.90	2.50	2.30	2.23	0.31	14
	Reference 2	3	1	< 1.5	2.80	1.90			
	Reference 3	2	0	1.70	2.30	2.00	2.00	0.42	21
	Reference 4	3	1	< 1.5	2.00	1.90			
	Reference Means					2.03			
	Study	10	1	< 1.5	3.20	1.90			
Cadmium	Reference 1	3	2	< 0.05	0.07	< 0.05			
	Reference 2	3	2	< 0.05	0.05	< 0.05			
	Reference 3	2	1	< 0.05	0.05	< 0.05			
	Reference 4	3	1	< 0.05	0.10	0.05			
	Reference Means								
	Study	10	3	< 0.05	0.10	0.05			
Copper	Reference 1	3	0	2.90	4.00	3.20	3.37	0.57	17
	Reference 2	3	0	3.10	5.80	5.30	4.73	1.44	30
	Reference 3	2	0	3.20	3.80	3.50	3.50	0.42	12
	Reference 4	3	0	4.20	5.10	4.70	4.67	0.45	10
	Reference Means					4.18	4.07		
	Study	10	0	2.90	4.80	3.90	3.94	0.63	16
Mercury	Reference 1	3	0	0.06	0.10	0.08	0.08	0.02	25
	Reference 2	3	0	0.06	0.10	0.07	0.08	0.02	27
	Reference 3	2	0	0.09	0.10	0.10	0.10	0.01	7
	Reference 4	3	0	0.09	0.11	0.10	0.10	0.01	10
	Reference Means					0.09	0.09		
	Study	10	0	0.05	0.15	0.09	0.09	0.03	30

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV %
Selenium	Reference 1	3	0	0.70	0.80	0.70	0.73	0.06	8
	Reference 2	3	0	0.50	0.80	0.80	0.70	0.17	25
	Reference 3	2	0	0.70	0.70	0.70	0.70	0.00	0
	Reference 4	3	0	0.70	0.80	0.80	0.77	0.06	8
	Reference Means					0.75	0.73		
	Study	10	0	0.50	0.80	0.60	0.66	0.11	16
Silver	Reference 1	3	0	0.14	0.26	0.18	0.19	0.06	32
	Reference 2	3	0	0.15	0.25	0.20	0.20	0.05	25
	Reference 3	2	0	0.16	0.16	0.16	0.16	0.00	0
	Reference 4	3	0	0.21	0.27	0.23	0.24	0.03	13
	Reference Means					0.19	0.20		
	Study	10	0	0.15	0.25	0.20	0.21	0.03	15
Strontium	Reference 1	3	0	5.20	10.00	9.30	8.17	2.59	32
	Reference 2	3	0	8.90	13.00	10.00	10.63	2.12	20
	Reference 3	2	0	6.20	15.00	10.60	10.60	6.22	59
	Reference 4	3	0	5.10	18.00	6.00	9.70	7.20	74
	Reference Means					8.98	9.78		
	Study	10	0	4.40	18.00	8.95	10.03	4.64	46
Zinc	Reference 1	3	0	31.00	32.00	31.00	31.33	0.58	2
	Reference 2	3	0	23.00	30.00	23.00	25.33	4.04	16
	Reference 3	2	0	27.00	30.00	28.50	28.50	2.12	7
	Reference 4	3	0	31.00	35.00	33.00	33.00	2.00	6
	Reference Means					28.88	29.54		
	Study	10	0	17.00	33.00	30.50	28.20	4.78	17
% Fat, Crude	Reference 1	2	1	< 0.5	0.70	0.60			
	Reference 2	3	0	0.50	1.90	1.10	1.17	0.70	60
	Reference 3	2	0	0.60	1.30	0.95	0.95	0.49	52
	Reference 4	3	0	0.60	0.70	0.60	0.63	0.06	9
	Reference Means					0.81			
	Study	10	2	< 0.5	1.40	0.70			
% Moisture	Reference 1	3	0	78.00	81.00	79.00	79.33	1.53	2
	Reference 2	3	0	80.00	85.00	81.00	82.00	2.65	3
	Reference 3	2	0	79.00	82.00	80.50	80.50	2.12	3
	Reference 4	3	0	78.00	80.00	78.00	78.67	1.15	1
	Reference Means					79.63	80.13		
	Study	10	0	80.00	85.00	81.00	81.70	1.77	2

Notes:
- Metal concentrations are in mg/kg dry wt
- Fat and moisture are in % wet wt
- CV = Coefficient of Variation (SD as % of mean)

Table 6-15 Comparison of Body Burden Values in Crab Leg Composites Among 2000 and 2004 Samples

Variable	2000	2004	
	Study Area (*n* = 4 composites)	Study Area (*n* = 10 composites)	Pooled References (*n* = 11 composites)
% moisture	79-81	80-85	78-85
% lipid/fat[1]	0.59-0.71	< 0.5-1.4	< 0.5-1.9
Arsenic	4.8-6.8	4.8-12	6.8-13
Boron	1.7-3.2	< 1.5-3.2	< 1.5-2.8
Cadmium	< 0.08	< 0.05-0.10	< 0.05-0.10
Copper	3.0-4.2	2.9-4.8	2.9-5.8
Mercury	0.08-0.10	0.05-0.15	0.06-0.11
Selenium	0.50-0.70	0.5-0.8	0.5-0.8
Silver	< 0.12	0.15-0.25	0.14-0.27
Strontium	6.2-9.8	4.4-18	5.1-18
Zinc	24-31	17-33	23-35

Notes:
- Metal concentrations are mg/kg dry wt
- Study Area sampling in 2004 occurred over a larger area than Study Area sampling in 2000
- [1] % lipid was measured in 2000 and % crude fat was measured in 2004. The two measures are comparable but EQL in 2000 was lower than in 2004 (0.1% versus 0.5%)

Variation of crab leg body burden variables, with one exception noted below, has been remarkably limited over both time and space. Table 6-15 provides ranges for frequently detected variables, which rarely varied by more than two-fold over all 25 composites analyzed in 2000 and 2004. That summary omits “matches” of values below EQL over time and space for many infrequently detected or undetected variables (e.g., several unlisted metals, HCs, PAHs). These results are evidence of the consistency of analytical results, often at concentrations close to EQL (where analytical error is high), over time or space. The one exception is silver, which was not detected in 2000 but which occurred at detectable concentrations in both the Study and Reference Areas in 2004.

Additional analyses comparing the Study Area and Reference Areas were performed on 2004 data for moisture, fat content and concentrations of eight metals (arsenic, boron, copper, mercury, selenium, silver, strontium, zinc). Concentrations of seven of these eight metals were positively correlated with each other and with the first Principal Component (Metals PC1) derived from those concentrations (Table 6-16). Strontium concentrations were negatively correlated with concentrations of most other metals and with PC1. In other words, strontium concentrations in claws were lower when concentrations of other metals were higher. Metals PC1 accounted for almost half the total covariance among variables and variance among samples. PC1 scores were used as a summary measure for further analyses, with higher scores indicating higher concentrations of all metals except strontium.

Table 6-16 Correlations (Parametric or Pearson *r*) Between Metal Concentrations in Crab Claw Composites and Principal Components (PC) Derived from those Concentrations

Metal	Correlation (*r*) with:		
	PC1	PC2	PC3
Zinc	**0.866**	0.014	0.179
Silver	**0.825**	0.045	–0.041
Mercury	**0.748**	0.298	0.357
Arsenic	**0.667**	0.279	–0.329
Selenium	**0.665**	**–0.553**	–0.218
Copper	**0.610**	–0.209	**–0.659**
Boron	0.330	**–0.683**	**0.567**
Strontium	**–0.542**	–0.485	–0.227
% variance	45.6	15.3	14.1

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

PC2 and PC3 each accounted for approximately 15% of the total variance and covariance, not much more than for an individual variable (i.e., with eight variables, each should account for 1/8 or 12.5% of total variance). These secondary PCs could reflect real but subtle differences in either availability or uptake of metals, but could also be artifacts of non-linearity in the relationships identified by PC1, or the limited number of significant digits and decimal places (never more than EQL). For example, the expected or predicted value of Metal A based on concentrations of the other seven metals might be 5.5, with the observed value reported as either 5 or 6 if EQL = 1. The expected and observed values agree to one significant digit, with the agreement reflected in PC1. However, the difference between either 5 or 6 and 5.5 (≈ 10%) is unexplained and nuisance variance, potentially reflected in PC2 and PC3 (a continuity problem that will occur whenever most concentrations are below 10 times EQL).

Moisture, fat content and metal concentrations (Metals PC1 and individual metal concentrations) did not differ among Reference Areas or between the Study Area and Reference Areas (Table 6-17; Figure 6-6 plots PC1 scores by area). In general, *p* values for the Among References and SR contrasts converged on $p = 0.5$, which will be the case whenever there are small or no differences among areas. *p* values for the Among References contrast were usually lower than for the SR contrast, suggesting that whatever small differences occurred were among Reference Areas rather than between the Study Area and Reference Areas.

Table 6-17 Results (*p*) of Nested ANOVA Comparing Body Burdens in Crab Claw Composites Among Areas

Variable	Contrast		
	Among References	Study versus Reference (SR)	
		Error=	
		Among References	Among composites
% moisture	0.173	0.251	0.069
% fat	0.193	0.638	0.497
Metals PC1	0.425	0.766	0.750
Arsenic	0.097	0.793	0.657
Boron	0.683	0.555	0.643
Copper	0.082	0.829	0.706
Mercury	0.599	0.523	0.571
Selenium	0.857	0.069	0.180
Silver	0.212	0.753	0.662
Strontium	0.916	0.785	0.904
Zinc	0.136	0.635	0.453

Notes: - Table 6-6 and Section 6.3 provide details on the nested ANOVA and contrasts
- $p \leq 0.05$ (in bold)

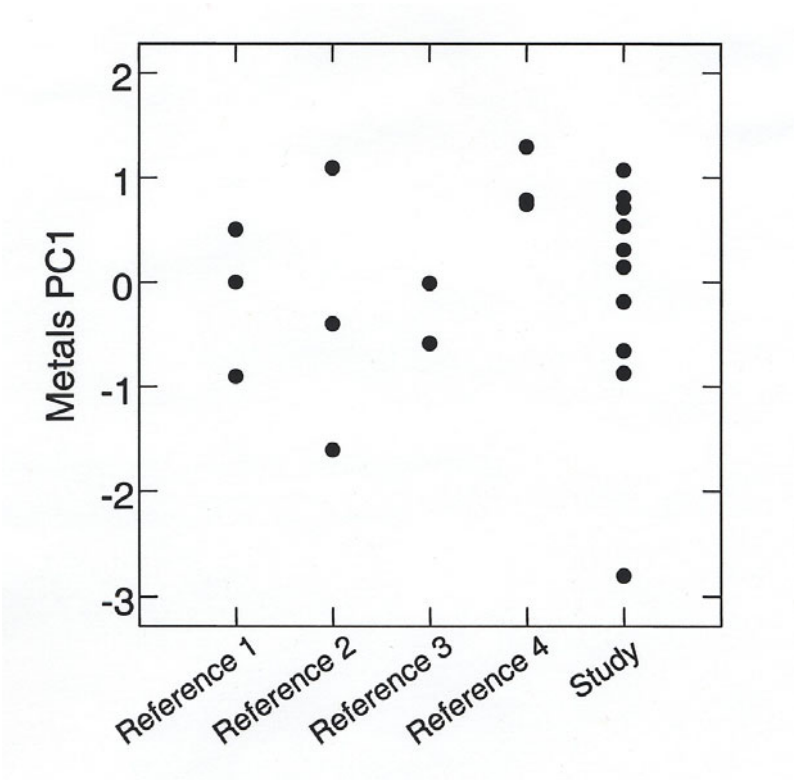


Figure 6-6 Distribution of Metals PC1 Scores for Crab Claws

Note: Some points may represent more than one composite sample

Spearman rank correlations (r_s) among body burden variables and between those variables and Biological Characteristics are provided in Table 6-18. Metals PC1 was used as a summary measure of concentrations of the eight metals, but correlations are also provided for mercury and strontium. Mercury is of interest because, as methyl mercury, it should accumulate and persist to a greater extent than other metals. Strontium is of interest because concentrations were negatively correlated with concentrations of other metals, and strontium should ultimately be incorporated into the shell or exoskeleton rather than edible tissue (e.g., claw meat).

Table 6-18 Spearman Rank Correlations (r_s) Among Crab Body Burden Variables, and Between Those Variables and Biological Characteristics

	% moisture	% fat	Metals PC1	Mercury	Strontium
Carapace width	–0.049	–0.184	0.329	0.406	–0.207
Claw height	0.001	–0.237	0.330	**0.514***	–0.168
% recent moult	0.218	0.255	–0.357	**–0.565****	0.429
% moisture		0.031	**–0.508***	–0.185	0.410
% fat			0.421	0.275	–0.072
Metals PC1				**0.717****	**–0.574****
Mercury					–0.037

Notes: - n = 21 composites from five areas
- *$p \leq 0.05$; **$p \leq 0.01$; r_s at $p \leq 0.05$ (in bold)

As expected, rank (non-parametric) correlations between mercury and strontium concentrations and Metals PC1 were similar to the parametric correlations given in Table 6-16, although concentrations of the two metals were uncorrelated rather than negatively correlated. Few other correlations were significant. Metals PC1 was negatively correlated with moisture content, indicating that dry weight metal concentrations decreased with increasing moisture content. That, in turn, indicates that differences in wet weight concentrations among composites would be greater than for dry weight concentrations.

Metals PC1 (i.e., metal concentrations) increased with increasing size and decreased with increasing % recent moult (Table 6-18). Correlations between mercury concentrations and the three size variables were similar and stronger. Therefore, metals, particularly mercury, may persist and biomagnify to some extent, with concentrations increasing with size and presumably age. 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). Concentrations of metals (except strontium) were also lower in recent moults, suggesting that some metals may be transferred from muscle to shell prior to moulting. However, the correlations between metal concentrations and % recent moult could be an artifact of correlations between the latter and size.

Correlations among body burden variables, and between those variables and Biological Characteristics, within Reference Areas and within the Study Area were similar to those provided in Table 6-18 for all areas combined. Therefore, the relationships between body burden and biological variables in Table 6-18 and discussed above were natural and unrelated to project activity.

6.4.2.2 Plaice

Liver

Summary statistics for moisture, fat content and concentrations of detected substances in plaice liver composites in 2004 are provided in Table 6-19. Comparison of 2004 summary statistics to 2000 values are provided in Table 6-20. In one Study Area sample in 2004, concentrations of compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} ranges were less than an EQL of 65 mg/kg. The EQL for other samples were 15 mg/kg, and many values greater than the EQL of 15 mg/kg were less than 65 mg/kg. Therefore, the values less than 65 mg/kg were deleted from summary tables and subsequent analysis.

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

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV%
$>C_{10}$-C_{21}	Reference 1	3	0	31	87	70	62.67	28.71	46
	Reference 2	3	0	44	56	49	49.67	6.03	12
	Reference 3	3	0	76	85	78	79.67	4.73	6
	Reference 4	3	0	110	150	140	133.33	20.82	16
	Reference Means					84.25	81.33		
	Study	9	0	47	110	65	74.33	24.66	33
$>C_{21}$-C_{32}	Reference 1	3	0	62	130	79	90.33	35.39	39
	Reference 2	3	0	64	110	71	81.67	24.79	30
	Reference 3	3	0	57	100	65	74.00	22.87	31
	Reference 4	3	0	56	96	91	81.00	21.79	27
	Reference Means					76.5	81.75		
	Study	9	0	40	120	55	62.11	23.29	37
$>C_{10}$-C_{32}	Reference 1	3	0	93	220	150	154.33	63.61	41
	Reference 2	3	0	120	150	120	130.00	17.32	13
	Reference 3	3	0	140	180	150	156.67	20.82	13
	Reference 4	3	0	200	250	200	216.67	28.87	13
	Reference Means					155	164.4		
	Study	10	1	< 65[1]	200	115			
Arsenic	Reference 1	3	0	2.8	4.3	2.9	3.33	0.84	25
	Reference 2	3	0	1.8	5.2	4	3.67	1.72	47
	Reference 3	3	0	3.1	3.4	3.1	3.20	0.17	5
	Reference 4	3	0	4.1	5.4	4.3	4.60	0.70	15
	Reference Means					3.58	3.7		
	Study	10	0	1.8	5.8	3.35	3.42	1.08	32

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV%
Cadmium	Reference 1	3	0	0.38	0.69	0.41	0.49	0.17	35
	Reference 2	3	0	0.46	0.65	0.48	0.53	0.10	20
	Reference 3	3	0	0.38	0.41	0.41	0.40	0.02	4
	Reference 4	3	0	0.49	0.65	0.53	0.56	0.08	15
	Reference Means					0.46	0.50		
	Study	10	0	0.33	0.54	0.435	0.44	0.07	17
Copper	Reference 1	3	0	3.1	4.9	4.2	4.07	0.91	22
	Reference 2	3	0	2.8	4.6	4.5	3.97	1.01	26
	Reference 3	3	0	3.3	4.7	4	4.00	0.70	18
	Reference 4	3	0	3	6.6	5.1	4.90	1.81	37
	Reference Means					4.45	4.23		
	Study	10	0	1.8	6	3.4	3.62	1.42	39
Iron	Reference 1	3	0	22	66	44	44.00	22.00	50
	Reference 2	3	0	36	58	52	48.67	11.37	23
	Reference 3	3	0	30	36	33	33.00	3.00	9
	Reference 4	3	0	32	45	42	39.67	6.81	17
	Reference Means					42.75	41.33		
	Study	10	0	29	52	41.5	40.50	7.82	19
Manganese	Reference 1	3	0	0.7	0.8	0.8	0.77	0.06	8
	Reference 2	3	0	0.8	0.9	0.8	0.83	0.06	7
	Reference 3	3	0	0.8	1	0.8	0.87	0.12	13
	Reference 4	3	0	0.7	1	0.9	0.87	0.15	18
	Reference Means					0.825	0.833		
	Study	10	0	0.7	1	0.8	0.83	0.09	11
Mercury	Reference 1	3	0	0.02	0.04	0.03	0.03	0.01	33
	Reference 2	3	0	0.03	0.04	0.04	0.04	0.01	16
	Reference 3	3	0	0.02	0.04	0.03	0.03	0.01	33
	Reference 4	3	0	0.03	0.04	0.03	0.03	0.01	17
	Reference Means					0.0325	0.033		
	Study	10	0	0.02	0.04	0.03	0.03	0.00	16
Selenium	Reference 1	3	0	1.7	2.1	1.9	1.90	0.20	11
	Reference 2	3	0	2.1	2.5	2.4	2.33	0.21	9
	Reference 3	3	0	1.9	2.2	2	2.03	0.15	8
	Reference 4	3	0	1.3	1.8	1.7	1.60	0.26	17
	Reference Means					2	1.97		
	Study	10	0	1.7	2.3	1.95	1.98	0.18	9
Silver	Reference 1	3	3	< 0.12	< 0.12	< 0.12			
	Reference 2	3	3	< 0.12	< 0.12	< 0.12			
	Reference 3	3	2	< 0.12	0.13	< 0.12			
	Reference 4	3	2	< 0.12	0.18	< 0.12			
	Reference Means								
	Study	10	10	< 0.12	< 0.12	< 0.12			

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV%
Strontium	Reference 1	3	3	< 1.5	< 1.5	< 1.5			
	Reference 2	3	3	< 1.5	< 1.5	< 1.5			
	Reference 3	3	3	< 1.5	< 1.5	< 1.5			
	Reference 4	3	2	< 1.5	1.6	< 1.5			
	Reference Means								
	Study	10	10	< 1.5	< 1.5	< 1.5			
Zinc	Reference 1	3	0	23	25	23	23.67	1.15	5
	Reference 2	3	0	23	24	24	23.67	0.58	2
	Reference 3	3	0	22	26	22	23.33	2.31	10
	Reference 4	3	0	22	29	28	26.33	3.79	14
	Reference Means					24.25	24.25		
	Study	10	0	19	24	22.5	22.20	1.75	8
% Fat, Crude	Reference 1	3	0	14	23	15	17.33	4.93	28
	Reference 2	3	0	11	13	12	12.00	1.00	8
	Reference 3	3	0	11	17	14	14.00	3.00	21
	Reference 4	3	0	15	18	16	16.33	1.53	9
	Reference Means					14.25	14.92		
	Study	10	0	10	20	12.5	13.30	2.87	22
% Moisture	Reference 1	3	0	63	70	68	67.00	3.61	5
	Reference 2	3	0	70	71	70	70.33	0.58	1
	Reference 3	3	0	66	71	68	68.33	2.52	4
	Reference 4	3	0	66	69	67	67.33	1.53	2
	Reference Means					68.25	68.25		
	Study	10	0	66	73	70	69.90	2.02	3

Notes:
- Metal and HC concentrations are in mg/kg dry wt
- Fat and moisture are in % wet wt
- CV = Coefficient of Variation (SD as % of mean)
- [1] EQL < 65 because of insufficient tissue volume

Table 6-20 Comparison of Body Burden Values in Plaice Liver Composites Between 2000 and 2004 Samples

Variable	2000	2004	
	Study Area (*n* = 3 composites)	Study Area (*n* = 10 composites)	Pooled References (*n* = 12 composites)
Arsenic	1.4-26	1.8-5.8	1.8-5.4
Cadmium	0.65-1.2	0.33-0.54	0.38-0.69
Copper	3.9-5.5	1.8-6.0	2.8-6.6
Iron	29-110	29-52	22-66
Manganese	< 1-1.1	0.7-1.0	0.7-1.0
Mercury	0.03-0.04	0.02-0.04	0.02-0.04
Selenium	1.9-3.0	1.7-2.3	1.3-2.5
Zinc	25-39	19-24	22-29

Notes:
- Metal concentrations are mg/kg dry wt
- Study Area sampling in 2004 occurred over a larger area than Study Area sampling in 2000

Values of most body burden variables in plaice liver did not differ substantially between 2000 (baseline) and 2004 (Table 6-20). Concentrations of some metals (e.g., arsenic, cadmium, iron) varied more within the Study Area in 2000 than in 2004, although one would normally expect a wider range with the larger sample sizes used in 2004. The same eight metals were frequently detected in both years (and the other metals and PAHs were rarely or never detected). Concentrations of compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} range were detected in 21 composites in 2004, but were not detected at the same EQL in any 2000 composites. However, PSC Maxxam Analytics (J. McDonald, pers. comm.) reports that these compounds were fatty acids rather than HCs originating from drill muds, fuel or lubricating oils.

Additional analyses were performed on 2004 moisture data, fat content, concentrations of eight metals (arsenic, cadmium, copper, iron, manganese, mercury, selenium, zinc) and concentrations of compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} range. Concentrations of six of the eight metals were positively correlated with each other and with the first Principal Component (Metals PC1) derived from those concentrations (Table 6-21). Correlations were strongest for arsenic, cadmium, copper and zinc. The other four metals were positively correlated with PC2. Thus, there appeared to be two groups of metals: the PC1 metals (arsenic, cadmium, copper, copper, zinc) and the PC2 metals (iron, manganese, mercury, selenium), although overall, correlations of most metals were positively correlated. PC1 and PC2 were retained for further analyses. PC3 was not, since it did not account for much more variance than a single original variable.

Table 6-21 Correlations (Parametric or Pearson *r*) Between Metal Concentrations in Plaice Liver Composites and Principal Components (PC) Derived from those Concentrations

Metal	Correlation (*r*) with:		
	PC1	PC2	PC3
Copper	**0.912**	–0.189	0.253
Zinc	**0.782**	–0.152	–0.100
Cadmium	**0.676**	0.306	**–0.573**
Arsenic	**0.601**	–0.418	0.023
Mercury	0.491	**0.620**	0.135
Manganese	0.356	0.476	**0.535**
Selenium	–0.152	**0.599**	0.431
Iron	–0.098	**0.673**	**–0.563**
% variance	31.1	21.9	15.1

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

Most *p* for comparisons of moisture, fat content and metal concentrations among areas were greater than 0.10 and many were greater than 0.5 (Table 6-22; Figure 6-7 also plots Metals PC1 and PC2 scores by area). Some results for individual metals may be suspect because only a few values near EQL were observed. For example, there were only three mercury values (0.02, 0.03,

0.04 mg/kg; EQL = 0.01 mg/kg) and only four manganese values (0.7, 0.8, 0.9. 1.0 mg/kg; EQL = 0.5 mg/kg).

Table 6-22 Results (*p*) of Nested ANOVA Comparing Body Burdens in Plaice Liver Composites Among Areas

Variable	Contrast		
	Among References	Study versus Reference (SR)	
		Error=	
		Among References	Among composites
% moisture	0.272	0.235	0.095
% fat	0.155	0.430	0.217
Metals PC1	0.243	0.206	0.063
Metals PC2	0.184	0.953	0.933
Arsenic	0.387	0.592	0.544
Cadmium	0.208	0.322	0.144
Copper	0.791	0.161	0.290
Iron	0.340	0.877	0.856
Manganese	0.579	0.931	0.938
Mercury	0.560	0.368	0.386
Selenium	**0.002**	0.957	0.875
Zinc	0.272	0.142	**0.031**
$>C_{10}$-C_{21} [1]	**0.001**	0.819	0.474
$>C_{21}$-C_{32} [1]	0.886	**0.031**	0.095

Notes:
- Table 6-6 and Section 6.3 provide details on the nested ANOVA and contrasts
- $p \leq 0.05$ (in bold)
- [1] One Study Area value < EQL of 65 mg/kg deleted

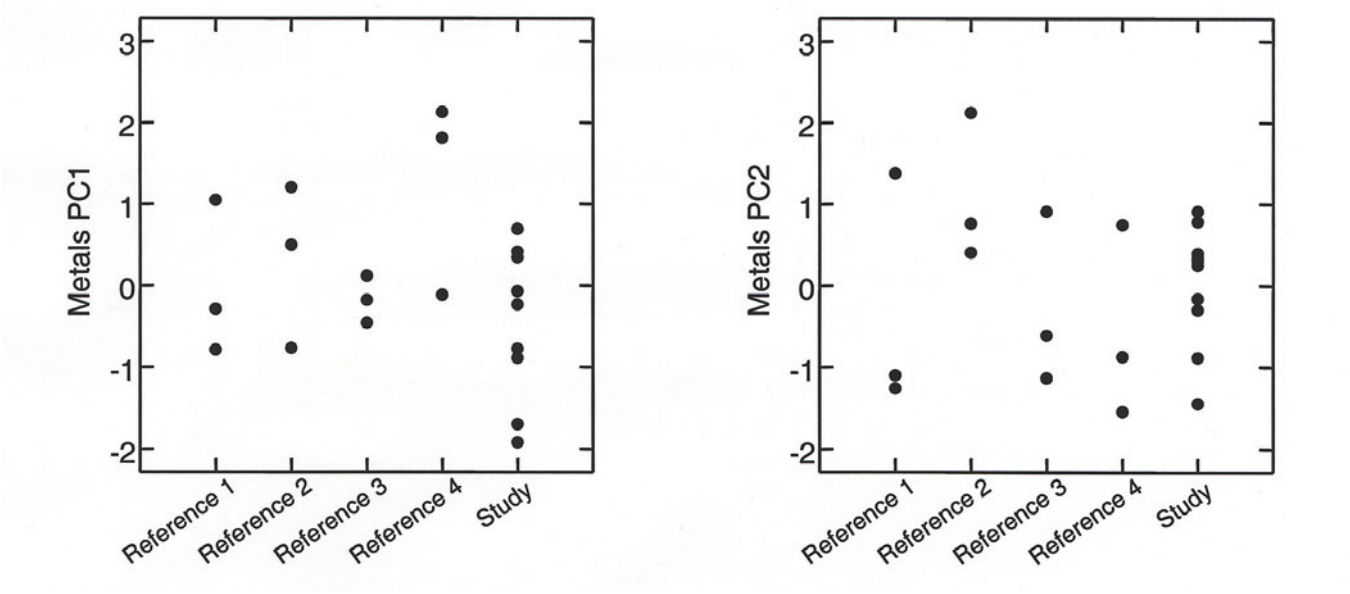


Figure 6-7 Distribution of Metals PC1 and PC2 Scores for Plaice Liver

Note: Some points may represent more than one sample

Selenium concentrations differed significantly among the Reference Areas (Table 6-22), with concentrations higher in Reference Area 2 and lower in Reference Area 4, than in the other two Reference Areas (and the Study Area) (Table 6-19). The differences were relatively small, since the overall range was only 1.3 to 2.5 mg/kg, or approximately two-fold. Zinc concentrations differed significantly between the Study and Reference Areas, if tested against the variance among composites (reasonable, given that $p > 0.20$ for the Among References contrast) (Table 6-22). Zinc concentrations were lower in the Study Area composites (Table 6-19). Metals PC1 scores (correlated with zinc concentrations) were also lower in the Study Area (Figure 6-7). Any differences in Metals PC2 scores were among Reference Areas, not between the Study and Reference Areas, and driven largely by differences in selenium concentrations (Table 6-22; Figure 6-7).

Concentrations of compounds in the $>C_{10}$-C_{21} range differed significantly among Reference Areas (Table 6-22) and were approximately twice as great in Reference Area 4 than in the other four areas (Table 6-19). In contrast, there were no differences in concentrations of compounds in the $>C_{21}$-C_{32} range among Reference Areas, but concentrations were lower in the Study Area than in the Reference Areas (Table 6-22; Table 6-19). Those results, and specifically the SR contrast, should be regarded with some suspicion. Except for a few low values below 50 mg/kg, Study Area values were within the range of Reference Area values (Figure 6-7). The SR contrast was not significant when tested against the variance among composites (Table 6-22; $0.05 < p < 0.10$, an admittedly ambiguous result). The SR contrast was significant when tested against the variance among Reference Areas only because that variance was small (MS(A{R} < MSE; $F < 1$: $p >> 0.5$; all of which indicate *negative* added variance among Reference Areas). This is a statistical anomaly that can occur in nested designs when sample sizes are limited at the first (highest) level of replication (i.e., Reference Areas), and variances at that level are poorly or imprecisely estimated. A safe conclusion is that lower concentrations of compounds in the $>C_{21}$-C_{32} range were somewhat more likely to occur in the Study Area than in other areas.

Spearman rank correlations among body burden variables, and between those variable and composite mean weight, are provided in Table 6-23. Selenium and zinc were included because they were the only individual metals to differ significantly among areas (Table 6-22). As expected, rank correlations between selenium and zinc, and the Metals PCs, were similar to the parametric correlations (r) in Table 6-21. The rank correlation between Metals PC1 and PC2 was 0.14, whereas the parametric correlation (r) must be 0 (i.e., PC are parametrically uncorrelated). The difference (0.14 versus 0) is a useful indicator of differences attributable to the (often arbitrary) choice of parametric versus non-parametric methods, analyses and transforms for these data. In other words, corresponding r for Table 6-21 might be $r_s \pm 0.1$ - 0.2, so $|r_s| \leq 0.2$ could be an artifact of the non-parametric method chosen.

Table 6-23 Spearman Rank Correlations (r_s) Among Plaice Liver Body Burden Variables, and Between Those Variables and Composite Mean Gutted Weight

	% moisture	% fat	Metals PC1	Metals PC2	Selenium	Zinc	>C_{10}-C_{21}	>C_{21}-C_{32}
Mean weight	0.078	–0.072	0.053	0.025	0.113	0.196	–0.083	–0.292
% moisture		**–0.964****	0.407	**0.627****	**0.662****	0.131	–0.274	**–0.480***
% fat			–0.309	**–0.591****	**–0.776****	–0.113	0.243	**0.455***
Metals PC1				0.140	–0.200	**0.647****	0.232	–0.137
Metals PC2					**0.629****	–0.135	–0.194	0.186
Selenium						0.113	**–0.503***	–0.181
Zinc							0.097	–0.029
>C_{10}-C_{21}								0.208

None of the body burden variables was correlated with mean weight of fish in composites, indicating that differences in mean weight among composites had little effect on results for body burden analyses. Moisture and fat content were strongly negatively correlated, a correlation expected in fatty tissue such as liver. In liver, fat content was 10 to 20% wet weight and moisture content was approximately 70% wet weight. Thus, fat plus moisture accounted for 80 to 90% of tissue wet weight. When two variables account for most of a total, they will be negatively correlated.

Metals PC1 and especially Metals PC2 were positively correlated with moisture content, indicating that dry weight metal concentrations increased with increasing moisture content. Those correlations, the opposite of correlations observed for crab claws (Section 6.4.2.1; Table 6-17) and, to some extent, plaice fillets (see below), suggest that differences in wet weight concentrations among liver composites were smaller than differences in dry weight concentrations. Metals PC1 and PC2 were *negatively* correlated with fat content, indicating that metal concentrations generally *decreased* with increasing fat content. Those correlations were probably an artifact of the strong negative correlation between moisture and fat content. Normally, metal accumulation and concentrations should not be a function of fat or lipid content. Exceptions might be methyl mercury and possibly some forms of arsenic or cadmium, which may occur at *higher* rather than *lower* concentrations in fattier tissue.

In contrast to metal concentrations, concentrations of compounds in the >C_{10}-C_{21} and >C_{21}-C_{32} ranges were negatively correlated with moisture content and positively correlated with fat content (Table 6-23). Concentrations in the >C_{10}-C_{21} and >C_{21}-C_{32} ranges were relatively uncorrelated, Overall, concentrations of these organic compounds were uncorrelated with metal concentrations, except that concentrations of compounds in the >C_{10}-C_{21} range were negatively correlated with selenium concentrations. That correlation was restricted to the Reference Areas; selenium concentrations tended to be high in composites with low concentrations of compounds in the >C_{10}-C_{21} range.

Correlations among body burden variables, and between those variables and composite mean gutted weight, within Reference Areas and within the Study Area, were usually similar in sign if not strength to the overall correlations in Table 6-23. The exception was the negative correlation between selenium and concentrations of compounds in the $>C_{10}$-C_{21} range, noted above, which was restricted to the Reference Areas.

Fillets

Summary statistics for moisture, fat contents and concentrations of metals detected in at least one plaice fillet composite are provided in Table 6-24. Comparison of body burden values in plaice fillet composites between 2000 and 2004 samples are provided in Table 6-25.

Table 6-24 Summary Statistics for Plaice Fillet Body Burden (2004)

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV%
Arsenic	Reference 1	3	0	1.9	2.9	2.6	2.47	0.51	21
	Reference 2	3	0	2.1	2.6	2.2	2.30	0.26	12
	Reference 3	3	0	2.6	3.5	3.3	3.13	0.47	15
	Reference 4	3	0	3.4	4	3.5	3.63	0.32	9
	Reference Means					2.9	2.88		
	Study	10	0	2	4.2	2.75	2.79	0.68	24
Iron	Reference 1	3	3	< 15	< 15	< 15			
	Reference 2	3	3	< 15	< 15	< 15			
	Reference 3	3	3	< 15	< 15	< 15			
	Reference 4	3	3	< 15	< 15	< 15			
	Reference Means								
	Study	10	9	< 15	38	<15			
Mercury	Reference 1	3	0	0.07	0.12	0.09	0.09	0.03	27
	Reference 2	3	0	0.07	0.1	0.09	0.09	0.02	18
	Reference 3	3	0	0.06	0.09	0.06	0.07	0.02	25
	Reference 4	3	0	0.05	0.08	0.08	0.07	0.02	25
	Reference Means					0.08	0.08		
	Study	10	0	0.04	0.1	0.09	0.08	0.02	25
Selenium	Reference 1	3	3	< 0.5	< 0.5	< 0.5			
	Reference 2	3	3	< 0.5	< 0.5	< 0.5			
	Reference 3	3	3	< 0.5	< 0.5	< 0.5			
	Reference 4	3	3	< 0.5	< 0.5	< 0.5			
	Reference Means								
	Study	10	9	< 0.5	0.5	< 0.5			
Strontium	Reference 1	3	3	< 1.5	< 1.5	< 1.5			
	Reference 2	3	3	< 1.5	< 1.5	< 1.5			
	Reference 3	3	2	< 1.5	1.5	< 1.5			
	Reference 4	3	3	< 1.5	< 1.5	< 1.5			
	Reference Means								
	Study	10	10	< 1.5	< 1.5	< 1.5			
Zinc	Reference 1	3	0	4.2	4.6	4.4	4.40	0.20	5
	Reference 2	3	0	4.3	4.8	4.3	4.47	0.29	6

Variable	Area	*n*	*n* < EQL	Min	Max	Median	Mean	SD	CV%
	Reference 3	3	0	4	4.3	4.2	4.17	0.15	4
	Reference 4	3	0	3.8	4	3.9	3.90	0.10	3
	Reference Means					4.2	4.23		
	Study	10	0	3.4	4.8	4.2	4.20	0.36	8
% Fat, Crude	Reference 1	3	0	1.1	1.9	1.7	1.57	0.42	27
	Reference 2	3	0	1.1	1.4	1.2	1.23	0.15	12
	Reference 3	3	0	2.2	3.6	2.5	2.77	0.74	27
	Reference 4	3	0	1.1	3.1	2.2	2.13	1.00	47
	Reference Means					1.9	1.93		
	Study	10	0	1	3.3	1.95	1.99	0.67	33
% Moisture	Reference 1	3	0	77	80	78	78.33	1.53	2
	Reference 2	3	0	77	79	78	78.00	1.00	1
	Reference 3	3	0	77	81	79	79.00	2.00	3
	Reference 4	3	0	80	81	81	80.67	0.58	1
	Reference Means					79	79		
	Study	10	0	78	81	78.5	78.90	1.10	1

Notes:
- Metal concentrations are in mg/kg dry wt
- Fat and moisture are in % wet wt
- CV = Coefficient of Variation (SD as % of mean)

Table 6-25 Comparison of Body Burden Values in Plaice Fillet Composites Between 2000 and 2004 Samples

Variable	2000	2004	
	Study Area (*n* = 5 composites)	Study Area (*n* = 10 composites)	Pooled References (*n* = 12 composites)
% moisture	80-83	78-81	77-81
% lipid/fat[1]	0.72-1.44	1.0-3.3	1.1-4.2
Arsenic	1.1-1.9	2.0-4.2	1.9-4.0
Mercury	0.04-0.07	0.04-0.10	0.05-0.12
Zinc	3.1-3.8	3.4-4.8	3.8-4.8

Notes:
- Metal concentrations are mg/kg dry wt
- Study Area sampling in 2004 occurred over a larger area than Study Area sampling in 2000
- [1] % lipid was measured in 2000, and % crude fat was measured in 2004. The values are directly comparable among years

Moisture content and concentrations of arsenic, mercury and zinc in plaice fillet composite samples from 2000 and 2004 varied little over time and space (Table 6-25). Table 6-25 underestimates the consistency of results, since it excludes the many metals, HCs and PAHs that were rarely or never detected. Lipid content was higher in 2004 than in 2000.

Additional analyses were performed on 2004 moisture data, fat content and concentrations of arsenic. mercury and zinc. Moisture and fat content, and arsenic, mercury and zinc concentrations in plaice fillets did not differ between the Study and Reference Areas (all $p > 0.5$; Table 6-26). Moisture and fat content, and arsenic concentrations, may have differed among Reference Areas ($0.01 < p < 0.10$). Specifically, moisture content was slightly higher in Reference Area 4, and fat

content and arsenic concentrations higher in Reference Areas 3 and 4, than in other Reference Areas (Table 6-24).

Table 6-26 Results (*p*) of Nested ANOVA Comparing Body Burden Variables in Plaice Fillet Composites Among Areas

Variable	Contrast		
	Among References	Study versus Reference (SR)	
		Error=	
		Among References	Among composites
% moisture	0.076	0.917	0.853
% fat	0.055	0.905	0.822
Arsenic	**0.036**	0.851	0.704
Mercury	0.390	0.756	0.729
Zinc	0.112	0.872	0.793

Notes: - Table 6-6 and Section 6.3 provide details on the nested ANOVA and contrasts
- $p \leq 0.05$ (in bold)

Body burden variables for plaice fillets were uncorrelated with composite mean gutted weight (Table 6-27), although there was some evidence that mercury concentrations increased with weight ($p < 0.10$ for the positive correlation). Moisture and fat content were uncorrelated. Mercury and zinc concentrations were positively correlated with each other, but uncorrelated with arsenic concentrations. Dry weight zinc, but not arsenic or mercury, concentrations were negatively correlated with moisture content. Correlations within the Study Area or pooled Reference Area were similar in sign, if not strength, to the overall correlations in Table 6-27, with one exception. Arsenic concentrations were positively correlated with mercury and zinc concentrations in the Study Area, but negatively correlated with concentrations of the two other metals in the Reference Areas.

Table 6-27 Spearman Rank Correlations (r_s) Among Plaice Fillet Body Burden Variables, and Between Those Variables and Composite Mean Gutted Weight

	% moisture	% fat	Arsenic	Mercury	Zinc
Mean weight	0.178	–0.108	–0.106	0.402	0.145
% moisture		–0.094	0.221	–0.077	**–0.579****
% fat			0.139	–0.275	–0.267
Arsenic				0.188	–0.186
Mercury					**0.504***

Notes: - n = 22 composites from five areas
- $*p \leq 0.05$; $**p \leq 0.01$; r_s at $p \leq 0.05$ (in bold)

6.4.3 Taste Tests

No significant difference was noted between plaice from the Study and Reference Areas in both the triangle and hedonic scaling tests. Panelists for the triangle test were successful in discriminating only 11 out of 24 samples. These results were not significant at $\alpha = 0.05$ (Appendix C-3). ANOVA statistics for hedonic scaling are provided in Table 6-28. The results were not significant ($p = 0.88$; α

= 0.05), and from the frequency histogram (Figure 6-8), samples from both the Reference and Study Area were assessed similarly for preference. From ancillary comments (Table 6-29 and 6-30, and Appendix C-3), there were no consistent comments identifying abnormal or foreign odour or taste.

Table 6-28 Analysis of Variance for 2004 Preference Evaluation by Hedonic Scaling of Plaice

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.08333333	1	0.08333333	0.024798	0.875561	4.051742
Within Groups	154.583333	46	3.36050725			
Total	154.666667	47				

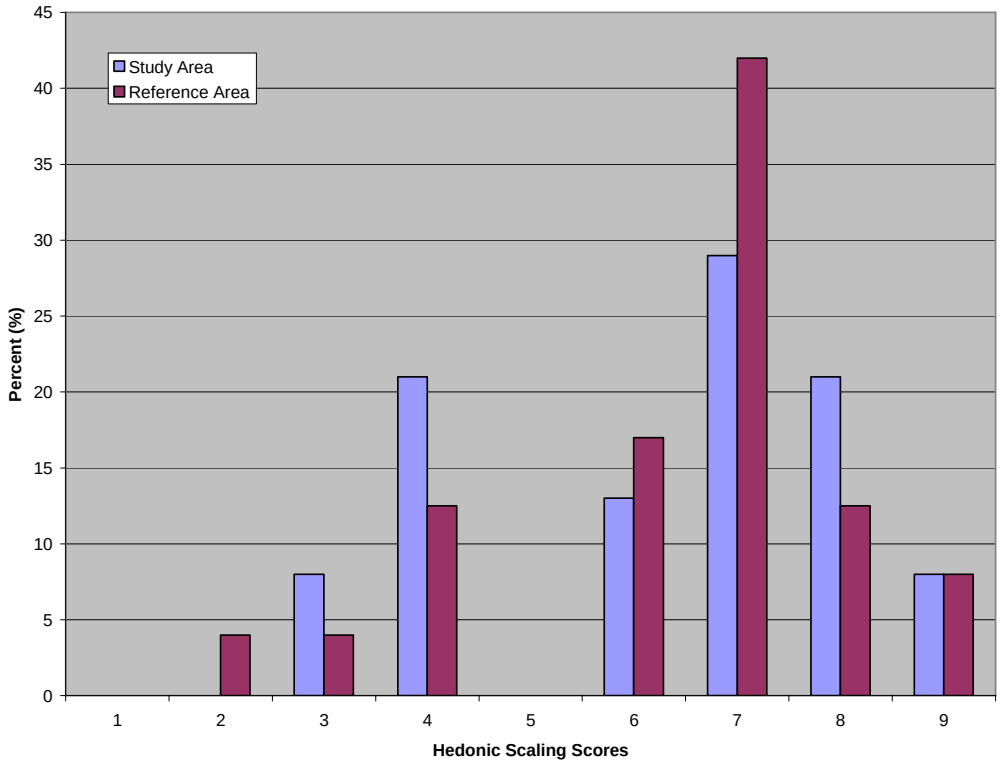


Figure 6-8 Plaice Frequency Histogram for Hedonic Scaling Sensory Evaluation (2004)

Table 6-29 Summary of Comments from the Triangle Test for Plaice (2004)

Reference Area (RA) Correctly Identified as Odd Sample	Study Area (SA) Correctly Identified as Odd Sample
I really could not detect much of a difference. 673 (RA) may have been a little different, but I would not be able to say why.	295 (SA) had a stronger odour and didn't taste as good as the other two. It was more dry as well.
Very difficult to tell, but 673 (RA) was slightly different. It had bones as well.	295 (SA) was better than the other two.
Odd one seemed to have a spice added.	413 (RA) white, mild taste, good scent. 549 (RA) very similar. 295 (SA) quite similar as well, somewhat brighter color, milder taste.
Sample 701 (RA) taste different and much drier.	Very hard to determine the difference, as there is really no noticeable difference.
Reference Area (RA) Incorrectly Identified as Odd Sample	**Study Area (SA) Incorrectly Identified as Odd Sample**
Quite difficult to say. All pretty much the same to me.	027 (SA) was much blander than the other two more flavourful samples. 673 (RA) & 164 (SA).
549 (RA) does not have a strong flavour as 413 (RA) and 295 (SA), also the odor is not as strong.	This sample was very wet. I prefer the drier sample.
641 (RA) better flavour.	027 (SA) had a stronger fish taste and was the firmest.
I have chosen 641 (RA) based on a very slight difference in odour (not flavour, texture, etc) however the difference is very slight.	Slightly stronger odour on 027 (SA) and 673 (RA) and texture different as well.
Taste off (almost like cod liver oil).	Unpleasant taste and odour.

Table 6-30 Summary of Comments from Hedonic Scaling Tests for Plaice (2004)

Preferred Reference Area (RA)	Preferred Study Area (SA)
412 (SA) had little texture and was "mushy", 382 (RA) had better flavour and texture.	382 (RA) little on bland side, taste more as if steamed without flavouring. 412 (SA) very tasty, steamed with flavour.
412 (SA) had a funny taste, sort of metallic maybe.	382 (RA) was much too watery and tasted very bland.
382 (RA) much more moist and a milder taste. 412 (SA) seems a little more oily. Sorry for not writing comments before.	Both tasted very similar. 967 (RA) bit drier.
Both tasted very similar. 967 (RA) bit drier.	967 (RA) had a weird grainy texture.
967 (RA) had a better texture and had a milder taste.	629 (SA), tasted better, had better odour, 967 (RA) looked nice, but had a strong taste.
967 (RA) had a perfect texture and a nice taste. 629 (SA) was nice but did not have as nice a texture (sort of wet).	Tastier than 532 (RA).
629 (SA) seems a bit mushy.	532 (RA) tasted fine but was full of bone. 590 (SA) has an unpleasant odour but tastes ok.
532 (RA) very pleasant tasting, pleasant scent, firm texture, good light color. 590 (SA) stronger scent, somewhat softer texture, good light color.	402 (RA) flesh is "drier" in texture and has an extra taste (may be burned).
Very strong taste not characteristic of the species.	No distinguishable taste difference. Liked both the same.
No distinguishable taste difference. Liked both the same.	Bad smell and taste on 402 (RA)
124 (SA) very strong flavour.	Both dry and unpleasant slightly bitter taste.
The 124 (SA) sample had a stronger "fishy" taste than 402 (RA).	
Both dry and unpleasant slightly bitter taste.	

For crab, panelists for the triangle test were successful in discriminating 13 out of 24 samples. These results are significant at $\alpha = 0.05$ (Appendix C-3). ANOVA statistics for hedonic scaling are provided in Table 6-31. These results were significant ($p = 0.02$; $\alpha = 0.05$) with a preference for samples from the Reference Areas (Figure 6-9). However, from ancillary comments (Table 6-32 and 6-33; Appendix C-3), there were no consistent comments identifying abnormal or foreign odour or taste for either the triangle or hedonic test. In addition to this, there is strong evidence that the crab offered to panelists was in poor condition both because of storage and thawing conditions and because crab were sampled in the summer, when bitter crab disease is prevalent. This, and additional interpretations of taste test results, is discussed further in Section 7.0.

Table 6-31 Analysis of Variance for 2004 Preference Evaluation by Hedonic Scaling of Crab

Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	8.33333333	1	8.33333333	5.2571429	0.026478	4.051742
Within Groups	72.9166667	46	1.58514493			
Total	81.25	47				

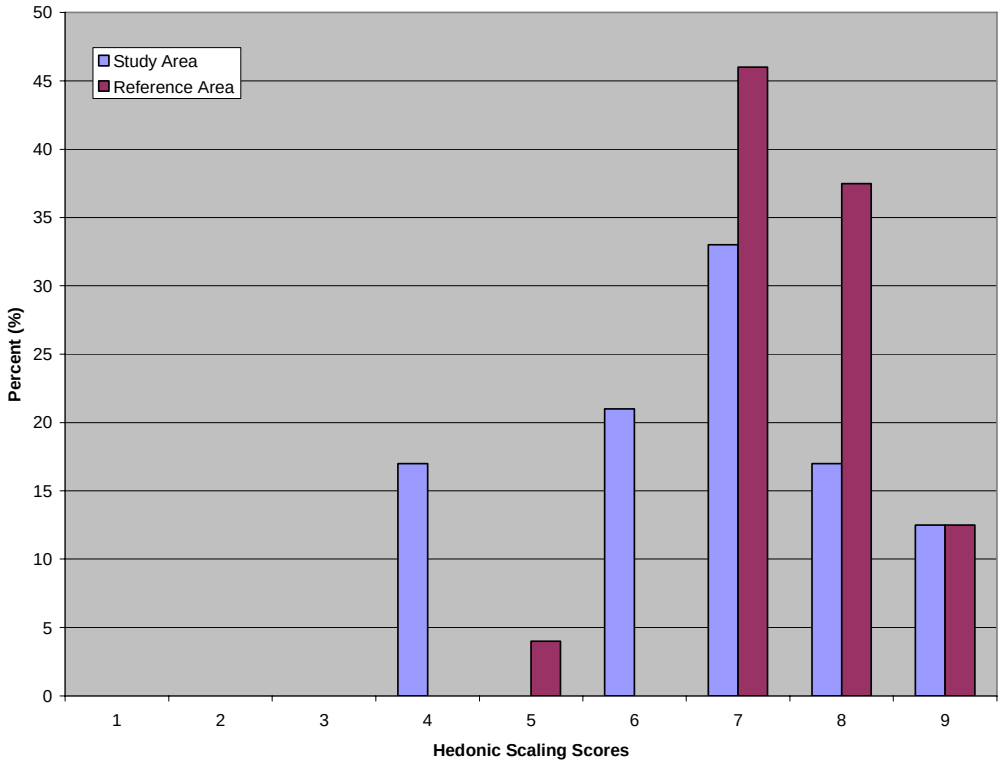


Figure 6-9 Crab Frequency Histogram for Hedonic Scaling Sensory Evaluation (2004)

Table 6-32 Summary of Comments from the Triangle Test for Crab (2004)

Reference Area (RA) Correctly Identified as Odd Sample	Study Area (SA) Correctly Identified as Odd Sample
371 (RA) had a slightly different odour but the taste was the same for all three.	The other two were sweeter.
Not as sweet as the other two.	634 (SA) did not have the same flavour as the other 2.
There was hard stuff in it.	All similar in texture and taste.
751 (RA) tasted sweeter and the other two were a little bitter.	Very slight difference between three samples. But the second one tasted different.
751 (RA) tasted better, not bitter.	
751 (RA) was sweeter, 427 (SA) & 908 (SA) had a bit of a bitter after taste.	
Reference Area (RA) Incorrectly Identified as Odd Sample	**Study Area (SA) Incorrectly Identified as Odd Sample**
No difference.	312 (SA) not as sweet and slight odour difference.
Much sweeter sample.	Seems a little more bitter? Not as sweet? That was hard!
Odd sample seemed to be a little dry.	Sample 908 (SA) a little more wetter than the other two, but still very tasty and likeable. All three are acceptable.
Very similar but 973 (RA) a little more flavour.	427 (SA) seemed sweeter.
973 (RA) is sweeter.	

Table 6-33 Summary of Comments from the Hedonic Scaling Test for Crab (2004)

Preferred Reference Area (RA)	Preferred Study Area (SA)
407 (RA) Texture and taste (sweetness) much better.	Not much difference between them.
Not much difference between them.	Not much difference in taste.
320 (SA) contained a lot of shoulder cartilage.	407 (RA) a little drier and slightly burnt flavour. But not really a big lot of difference.
Not much difference in taste.	608 (RA) flavour fine but a little grit is off putting. 105 (SA) flavour good and no grit.
407 (RA) a little more crab flavour.	Very little difference in these. 105 (SA) may be a little sweeter.
236 (RA) better texture and more juicy.	I'll buy either…
236 (RA) is a lot firmer, where 826 (SA) seems like it got a lot more water in it.	105 (SA) had a slightly sweeter flavour. 608 (RA) did not.
826 (SA) a little too sweet.	Sample 358 (SA) had a sweeter taste.
236 (RA) – more sweet than 826 (SA). Texture is similar in both. 826 (SA) has less crab flavour.	Both samples were very good, nice and sweet.
105 (SA) has a bitter taste on it.	Tasted equally good!
I'll buy either….	
Both samples were very good, nice and sweet.	
358 (SA) off taste. 810 (RA) little gritty	
358 (SA) tastes slightly bitter.	
Tasted equally good!	

6.4.4 Fish Health Indicators

The full report on plaice health indicators is provided in Appendix C-4. Highlights of results are provided below.

6.4.4.1 MFO Activity

MFO enzyme activities were analyzed separately in immature and spent female plaice (Figures 6-10 and 6-11). Although sample sizes were small, enzyme activities were also included for males (all maturity stages pooled) (Figure 6-12).

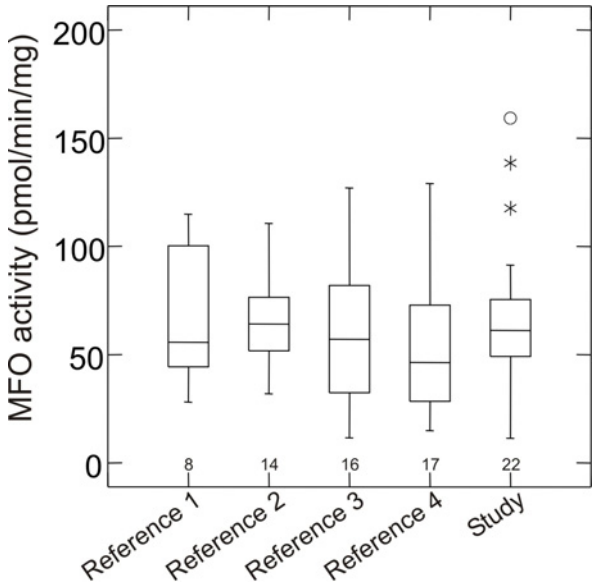


Figure 6-10 MFO Activity in Immature Females

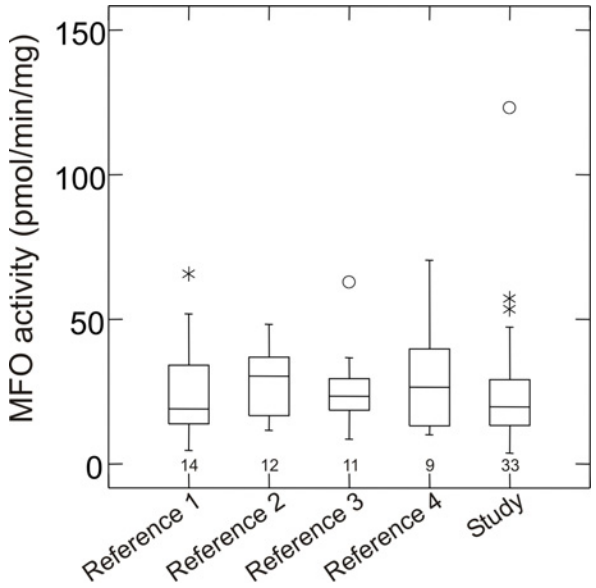


Figure 6-11 MFO Activity in Spent Females

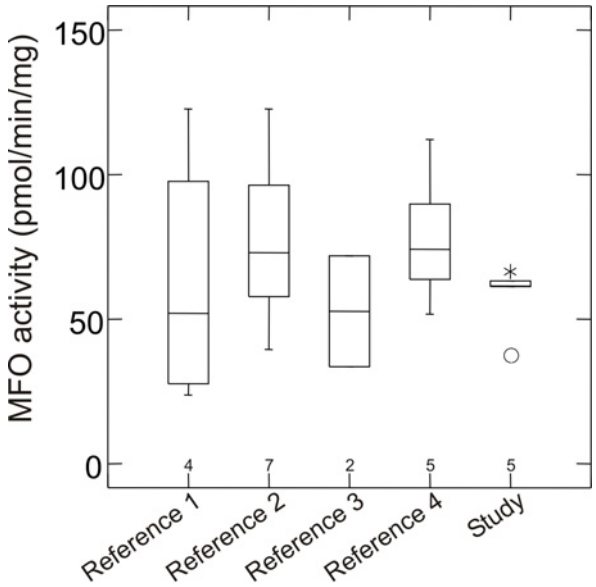


Figure 6-12 MFO Activity in Males

Notes:
- Horizontal line in middle of box = median. Box = 25th to 75th percentile
- Vertical lines = whiskers; include all values within 1.5 Hspread (75th minus 25th percentiles).
- The number under each box is the sample size
- Asterisks are outside values, > 1.5 Hpsreads from the 25th or 75th percentiles
- Circles are far outside values, > 3 Hspreads from the 25th or 75th percentiles

MFO enzyme activities did not differ significantly among Reference Areas or between the Study and Reference Areas, regardless of whether variance among areas or variance among fish was used as the error term, for any of the three groups (Nested ANOVA; Table 6-34).

Table 6-34 Results of Nested ANOVA Comparing MFO Activity Among Areas

Group	*p* values		
	Among References	Study versus Reference	
		Error=MS(A{R}) [1]	Error=MSE [2]
All males	0.631	0.460	0.525
Immature females	0.685	0.395	0.487
Spent females [3]	0.870	0.091	0.235

Notes:
- [1] Variance among Reference Areas used as the error term
- [2] Variance among fish within Areas used as the error term
- [3] MFO activity was log-transformed to reduce the effects of one high value

6.4.4.2 Gross Pathology

One fish from the Study Area had a tumour/cyst on the head kidney, while three fish from the Reference Areas displayed gill achromasia (or white gill) (Appendix C-4, Annex G, Photo 1).

6.4.4.3 Haematology

Blood smears collected in 2004 were considered of insufficient uniformity for carrying out reliable differential cell counts. Preliminary screening of the smears prepared in 2004 indicated that counts could vary by ± 20% or more upon examination of different regions of a slide. In human haematology, when 200 cells are counted, the variability is normally in the range of ± 7-10% (Lynch et al. 1969). Oceans Ltd. considers the variability found in the fish in 2004 too high for robust analysis.

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. Furthermore, greater accuracy can be obtained by this method through smearing a measurable amount of blood on each slide. This method has been used with fish by others on occasion (Blaxhall 1972; Arnold 2003) and has also been verified at the Oceans Ltd. laboratory.

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 6-35. The complete data set is provided in Appendix C-4 (Annex E) and representative photographs are included in Appendix C-4 (Annex G). Photos 2 and 6 (Appendix C-4, Annex G) represent a normal liver structure.

Table 6-35 Number of Plaice with Specific Types of Hepatic Lesions and Prevalence of Lesions in the 2004 White Rose Survey

Variable		Area					
		Reference 1	Reference 2	Reference 3	Reference 4	Reference Total	Study
No. fish		26	33	29	31	119	61
Nuclear pleomorphism	No.	0	0	0	0	0	1
	%	0.0	0.0	0.0	0.0	0.0	1.6
Megalocytic hepatosis	No.	0	0	0	0	0	1
	%	0.0	0.0	0.0	0.0	0.0	1.6
Eosinophilic foci	No.	0	0	0	0	0	1
	%	0.0	0.0	0.0	0.0	0.0	1.6
Basophilic foci	No.	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
Carcinoma	No.	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
Cholangio-fibrosis	No.	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0
Hydropic vacuolation	No.	0	0	0	0	0	0
	%	0.0	0.0	0.0	0.0	0.0	0.0
Macrophage aggregation [1]	No.	0	0	0	0	0	1
	%	0.0	0.0	0.0	0.0	0.0	1.6
Inflammatory response	No.	0	2	0	0	2	2
	%	0.0	6.1	0.0	0.0	1.7	3.3
Hepatocellular vacuolation	No.	0	3	3	3	9	2
	%	0.0	9.1	10.3	9.7	7.6	3.3
Biliary parasites	No.	6	10	6	3	25	16
	%	23.1	30.3	20.7	9.7	21.0	26.2

Note: - [1] Moderate to high aggregation (≥ 3 on a 0-7 relative scale)

Sixty-one fish from the Study Area and 119 fish from the four Reference Areas were examined and no cases of basophilic foci, clear cell foci, carcinoma, cholangioma, cholangiofibrosis or hydropic vacuolation were observed.

One case of nuclear pleomorphism associated with mild megalocytic hepatosis and moderate macrophage aggregation (Appendix C-4, Annex G, Photo 3), as well as one case of eosinophilic foci (Appendix C-4, Annex G, Photo 4), were noted from the Study Area.

Except for the case of moderate macrophage aggregation cited above, the frequencies of such aggregates in livers of fish from the different areas were very low.

Two fish from the Study Area and two fish from Reference Area 2 exhibited an inflammatory response (Appendix C-4, Annex G, Photo 5).

Nine fish from the Reference Areas and two fish from the Study Area showed a “patchy distribution” of hepatocellular vacuolation. These were all females, including 11 spent and (the only) one partly spent female. This type of vacuolation (Appendix C-4, Annex G, Photo 7) is likely a reflection of gonadal maturational stage. Incidences of hepatic vacuolation did not differ significantly among all five areas (G test p = 0.42), among the four Reference Areas (G test p = 0.69), or between the Study Area and the pooled Reference Areas (Fisher’s Exact Test p = 0.22). Except for the Study versus pooled Reference comparison, these tests and p are approximate, because of the low incidence of hepatic vacuolation in all areas.

An infestation of the biliary system with a myxosporean parasite (Appendix C-4, Annex G, Photo 8), possibly *Myxidium sp*., was found in 10 to 30% of the fish, with lower incidence in Reference Area 4 than in the other four areas (Table 6-35). Incidences of biliary parasites did not differ significantly among all five areas (G test p = 0.28), among the four Reference Areas (G test p = 0.21), or between the Study Area and the pooled Reference Areas (Fisher’s Exact Test p = 0.46).

One fish from Reference Area 2 exhibited a cluster of cells, identified as X-cells, in the liver (Appendix C-4, Annex G, Photo 9).

The observations on parasitism and X-cells 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

Seven fish (5.9%) from the Reference Areas (three from Reference Area 1, one from Reference Area 2 and three from Reference Area 4), including the three fish with achromasia, and one fish (1.6%) from the Study Area displayed extensive proliferation of ovoid and pale staining cells, or X-cells, in the interlamellar spaces of secondary lamellae (Appendix C-4, Annex G, Photo 10). 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 112 fish from the four Reference Areas and 60 fish from the Study Area (Table 6-36). The complete data set on fish from the 2004 survey is provided in Appendix C-4 (Annex F).

Table 6-36 Occurrence of Different Stages and Oedema Condition in the Gill of Plaice from the 2004 White Rose Survey

Variable	Area					
	Reference 1	Reference 2	Reference 3	Reference 4	All References	Study
No. fish	23	32	29	28	112	60
Stage 1: Thin lamellae[1]	49.7 ± 30.7	62.5 ± 20.5	55.9 ± 25.0	57.8 ± 22.1	56.5	53.2 ± 21.9
Stage 2: Distal hyperplasia[1]	32.0 ± 23.0	21.1 ± 17.2	24.9 ± 16.4	24.5 ± 15.8	25.6	30.3 ± 20.5
Stage 3: Epithelial lifting[1]	0.0	0.0	0.0	0.0	0.0	0.0
Stage 4a: Tip hyperplasia[1]	18.2 ± 19.3	16.4 ± 13.2	19.2 ± 16.2	17.6 ± 16.8	17.9	16.6 ± 14.7
Stage 4b: Telangiectasis[1]	0.03 ± 0.16	0.00	0.00	0.07 ± 0.24	0.02	0.00
Stage 5: Basal hyperplasia[1]	22.4 ± 21.9	16.9 ± 18.6	23.0 ± 24.8	24.6 ± 30.6	21.7	27.7 ± 24.6
Stage 6: Fusion[1]	0.00 ± 0.00	0.04 ± 0.25	0.00	0.00	0.01	0.00
Oedema condition[2]	1.10 ± 0.69	0.88 ± 0.46	1.19 ± 0.67	1.10 ± 0.68	1.07	0.93 ± 0.53

Notes:
- All data are expressed as mean ± standard deviation
- All References denotes means of the four Reference Area means
- Occurrences of stages and oedema condition are based on analysis of 100-120 lamellae per fish
- [1] Mean percentage ± SD of lamellae presenting the stage (in relation to the total number of lamellae counted)
- [2] Mean ± SD of rating on a 0-3 relative qualitative scale

Epithelial layers of secondary lamellae may vary in thickness. All the fish studied displayed a variable percentage of thin secondary lamellae, which is 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 layer (Appendix C-4, Annex G, Photo 11).

Distal hyperplasia (Appendix C-4, Annex G, Photo 12), tip hyperplasia (Appendix C-4, Annex G, Photo 13) or basal hyperplasia (Appendix C-4, Annex G, Photo 14) of secondary lamellae were observed in most of gill samples, indicating general lamellar thickening putatively of a background nature.

There were no significant differences in the occurrence of stages 1 to 5 and extent of oedema, after rank-transformation, among Reference Areas or between the Study and Reference Areas (Table 6-37); stages 3, 4b and 6 being excluded because they were rare or absent.

Table 6-37 Results of Nested ANOVA Comparing Some Gill Histopathology Variables Among Areas

Variable	*p* values		
	Among References	Study versus Reference	
		Error=MS(A{R}) [1]	Error=MSE [2]
Stage 1	0.536	0.267	0.247
Stage 2	0.281	0.259	0.117
Stage 4a	0.905	0.599	0.800
Stage 5	0.737	0.065	0.066
Oedema	0.320	0.472	0.374

Notes: - [1] Variance among Reference Areas used as the error term
- [2] Variance among fish within Areas used as the error term
- % occurrence of Stages 1-5, and extent of oedema, were rank-transformed

Microstructural changes, which have been associated with chemical toxicity, were absent or rarely observed. Fusion was seen in only one fish collected in Reference Area 2, while very mild telangiectasis was observed in one fish from Reference Area 1, and two fish from Reference Area 4. No cases of epithelial lifting were observed in either area. The levels of oedema (rated on a 0 to 3 relative scale) were quite low in all areas.

6.5 Key Findings

6.5.1 Biological Characteristics of Crab and Plaice

6.5.1.1 Crab

Crab size (carapace and claw size) differed significantly among composites within Reference Areas, and specifically within Reference Areas 1 to 3. Those differences represent small-scale spatial differences among trawls conducted in restricted areas.

Crab size was largest in Reference Area 4. Study Area crab were larger than crab in Reference Areas 1 and 2, and similar in size to Reference Area 3 crab.

Only one (of 32) crab in Reference 4 appeared to have moulted recently (i.e., in 2004), whereas 36 to 86% of crab in other areas were recent moults.

Smaller crab were more likely to have moulted recently.

Biological Characteristics of crab differed mostly among Reference Areas (and specifically between Reference Areas 1 to 3 and Reference Area 4) and there were no significant differences in Biological Characteristics, overall, between the Study and Reference Areas.

6.5.1.2 Plaice

Plaice liver and fillet body burden composites consisted of a mix of smaller immature and larger mature fish. Consequently, size distributions within composites were usually bimodal and truncated at the low end (since fish smaller than 250 mm in length were released). Therefore, a comparison of size within composites was not conducted.

Mean size (weight) of plaice in composites from Reference Area 1 and from the Study Area was greater than mean size for Reference Areas 2 to 4. However, differences among Reference Areas and between the Study and Reference Areas were not significant at $p \leq 0.05$. More extensive analyses of size and other Biological Characteristics broken down by sex and maturity status also revealed few or no biological differences among areas.

6.5.2 Body Burden

6.5.2.1 Crab

Moisture, fat content, and metal concentrations in crab claws in 2004 were similar to those measured in the Study Area in 2000. Differences within areas, among areas, and among years were rarely greater than two-fold.

HCs were not detected in any crab claw composite in either year.

Concentrations of seven of the eight frequently detected metals in crab claws (arsenic, boron, copper, mercury, selenium, silver, zinc) in 2004 were positively correlated with each other and negatively correlated with strontium concentrations.

Moisture and fat content, and metal concentrations, did not differ significantly among Reference Areas or between the Study Area and the Reference Areas.

Dry weight concentrations of metals (except strontium) were negatively correlated with moisture content. Concentrations of those metals, and especially mercury, were also positively correlated with crab size.

6.5.2.2 Plaice

Liver

Moisture content, fat content and metal concentrations in liver from the Study Area in 2004 were generally similar to values from 2000. However, fatty acids in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} ranges were detected in nine (of 10) Study Area liver composites and in all 12 Reference Area liver composites

in 2004. Fatty acids or other compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} ranges were not detected in fillet samples in 2000 or 2004. PAHs were not detected in any liver or fillet samples in either year.

The frequently detected metals in liver in 2004 fell into two groups: arsenic, cadmium, copper and zinc; and iron, manganese, mercury and selenium. However, there was also an overall tendency for concentrations of most metals to be positively correlated.

Moisture and fat content, and concentrations of most metals in liver did not differ significantly among Reference Areas or between the Study Area and Reference Areas. However, there were highly significant differences in concentrations of selenium and $>C_{10}$-C_{21} among Reference Areas. Concentrations of selenium were lowest and concentrations of $>C_{10}$-C_{21} were highest in Reference Area 4. Concentrations of zinc and $>C_{21}$-C_{32} were lower in the Study Area than in the Reference Areas, although the significance of those differences was dependant the statistical test used.

Dry weight metal concentrations in liver increased with decreasing moisture content and increasing fat content, whereas the reverse was true for $>C_{10}$-C_{21} and $>C_{21}$-C_{32}. Metal, $>C_{10}$-C_{21} and $>C_{21}$-C_{32} concentrations were largely uncorrelated. Size (i.e., mean weight of fish within composites) was uncorrelated with body burden variables.

Fillet

Moisture and fat content and concentrations of frequently detected metals in fillets collected in 2004 were similar to those in fillets collected from the Study Area in 2000.

Arsenic, mercury and zinc were detected in all fillet samples in 2004; other metals were not detected or were detected in only one or two (of 22) composites. Concentrations of these three metals, and moisture and fat content, did not differ between the Study Area and the Reference Areas (all $p > 0.5$). Differences among Reference Areas were greater, and significant for arsenic, with highest arsenic levels in Reference Area 4.

Fillet body burden variables were largely uncorrelated with each other and with fish size. However, zinc concentrations were positively correlated with mercury concentrations and negatively correlated with moisture content.

6.5.3 Taste Tests

There was no difference in taste between the Study and Reference Areas for plaice. However panelists were able to distinguish between Study and Reference Area crab and preferred Reference Area crab. However, there were no consistent comments resulting from the crab taste tests that identified abnormal or foreign odour or taste which would normally be associated with taint.

6.5.4 Fish Health Indicators

There were no significant differences in MFO enzyme activities in spent females, immature females or males among Reference Areas or between the Study and Reference Areas. With respect to gross pathology, one fish from the Study Area exhibited a tumour/cyst on the head kidney and three fish from the Reference Areas displayed gill achromasia (pale gill filaments). For tissue histopathology, one case of nuclear pleomorphism associated with megalocytic hepatosis and one case of oesinophilic foci were observed in liver tissues of fish from the Study Area. Liver tissue from some fish contained myxosporean parasites but no differences among Reference Areas or between the Study and Reference Areas were found. Liver lesions associated with chemical toxicity were generally absent or found only at very low incidence. For gills, a slightly higher percentage of basal hyperplasia, putatively of a background nature, was noted in the Study Area. Microstructural changes which have been associated with chemical toxicity and which could be more pathological in nature were absent or found at very low frequencies.

7.0 Discussion

7.1 Sediment Component

Evidence of project effects, particularly from recent drilling, in the White Rose EEM program can come from:

- changes in relationships between SQT variables and distances from drill centres between 2000 and 2004; and/or
- correlations between biological responses and physical or chemical alterations from drilling activity.

7.1.1 Physical and Chemical Characteristics

Sediments in the White Rose sampling area were uniformly sandy, with low gravel and fines content. The fines content in 2004 ranged from 1 to 3% and was similar to that in the nearby Terra Nova development (Petro-Canada 2003). Gravel content was lower and less variable in the White Rose sampling area than at Terra Nova.

The TOC content of sediments at White Rose was also low (less than 1.2 g/kg or 0.12 %). TOC values of 1% are considered typical of uncontaminated marine sediments (CCME 2005). In 2004, TOC was not strongly associated with fine particles. The correlation between TOC and fines content was slightly stronger in 2000 (Husky Energy 2001) but correlations for both years were weaker than at Terra Nova (Petro-Canada 2003).

In 2004, concentrations of $>C_{10}$-C_{21} HCs were elevated near the Southern drill centre and, to a lesser degree, near the Northern drill centre. Maximum concentrations at each drill centre were detected at station S5 (275 mg/kg) and N3 (28 mg/kg), located 300 m and 600 m from the Southern and Northern drill centres, respectively. Concentrations decreased with distance from both drill centres. In 2004, the median concentration of $>C_{10}$-C_{21} HCs was 22 mg/kg within 1 km of the Northern and Southern drill centres, and levels fell to approximately 1 mg/kg at distances of 5 km from these drill centres. Chromatograms for approximately 75% of the stations sampled within 8 km of the Northern or Southern drill centres had UCMs in the range of PureDrill IA-35. Low levels of $>C_{10}$-C_{21} HCs were detected at three stations located more than 8 km from the drill centres (stations 11, 12, and 27; HC range: 0.42 to 0.66 mg/kg). However, these HCs did not have UCMs in the range of Puredrill IA-35 and PSC Maxxam reports that these HCs are probably non-petrogenic material.

In 2004, barium concentrations were also elevated near the Southern drill centre and again, to a lesser degree, near the Northern drill centre. Maximum barium concentrations occurred at station

S5 (1,400 mg/kg) and concentrations decreased with distance from the Northern and Southern drill centres. Background levels of barium (less than 200 mg/kg) were reached within 2 km of the two drill centres, although very low-level contamination from drilling may have extended beyond that distance. There was some evidence that barium concentrations were naturally higher nearer the centre of the development and to the north than at other stations (Husky Energy 2001; comparison between years in this report). There were also significant positive correlations between concentrations of barium and other metals in 2000 and in 2004.

Directional effects were noted for both $>C_{10}$-C_{21} HCs and barium in 2004, with dispersion primarily to the southeast.

Concentrations of $>C_{10}$-C_{21} HCs and barium were excellent indicators of drilling activity for the White Rose development. The high concentrations observed near the Northern and Southern drill centres and the attenuation of those concentrations with distance provided unequivocal evidence of contamination from drilling activity. Higher concentrations near the Southern drill centre are consistent with drilling intensity with both SBMs and WBMs at this centre relative to the Northern drill centre from 2003 to 2004. The absence of elevated levels at the Central drill centre likely reflects the fact that drilling was limited at that centre and that SBMs had not been used there prior to EEM program sampling.

Elevated concentrations of $>C_{10}$-C_{21} HCs and barium have been observed near drill centres and platforms at other offshore oil developments and levels noted at White Rose were within the range of levels noted at these other sites (Table 7-1).

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[1] (mg/kg)
White Rose	2004	300 to 750 750 to 2500 2500 to 5000	8.74 to 275.92 0.21 to 21.95 <3 to 6.60	190 to 1400 120 to 470 140 to 230
	2000	300 to 750 750 to 2500 2500 to 5000	<3 <3 <3	140 to 180 140 to 190 140 to 210
Terra Nova	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

Well Location	Year of Study	Distance from Source (m)	Total Petroleum Hydrocarbon (mg/kg)	Barium[1] (mg/kg)
Hibernia (total extractable hydrocarbon data for 2000) (HMDC 2001)	2000	250 500 2,000	472 to 1,086 26 to 90 5 to 13	728 to 1,700 177 to 447 133 to 423
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	200 500 3,000	40 43 49	1,625 1,134 1,072
Gulf of Mexico (MU-A85) (Kennicutt et al. 1996)	1993	200 500 3,000	42.3 31.7 27.1	3,706 1,817 1,094
Gulf of Mexico (HI-A389) (Kennicutt et al. 1996)	1993	200 500 3,000	65 33 32	13,756 3,993 1,293
North Sea (Beatrice) (Addy et al. 1984)	1982	250 750 3,000	8 to 759 5 to 105 3 to 73	-
Dutch Continental Shelf (K14-13) (Daan and Mulder 1996)		200	54 to 161	-
Norway (Valhall) (Hartley 1996)	1985	250 500 3,000	-	19,000 to 96,000 3,700 to 9,300 280 to 430
North Sea (Brent) (Massie et al. 1985)	1981	800 3,200	41 to 61 33 to 43	-
North Sea (Forties) (Massie et al. 1985)	1980	800 3,200	9 to 78 16 to 55	-
Gulf of Mexico (Matagorda 622) (Chapman et al. 1991; Brooks et al. 1990)	1987	25 150 750 3,000	757 ±1,818	6,233 12,333 980
Santa Maria Basin (Hidalgo) (Phillips et al. 1998)	1991	125 500 1,000	-	1,250 975 1,050
Norway (Ekofisk) (Ellis and Schneider 1997)	1996	750 2,000 5,000	-	3,650 2,214 667
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 >500 to 2,000 >2,000 to 5,000	124 to 11,983 3 to 164 3 to 76	84 to 2,040 7 to 1595 8 to 729

Note: - [1] Absolute barium levels should not be compared across projects because of potential difference in measurement techniques (Hartley 1996); and differences in background levels.

Drilling activity may also have elevated fines content and sulphur concentrations within 1 km of the Southern drill centre. There was no relationship between fines content and distance from the Southern drill centre in 2000, but in 2004, fines content decreased with distance. Sulphur was not

measured in 2000, but the decrease in sulphur with distance is consistent with that of other drill mud indicators. There was less evidence for alteration in fines and sulphur content near the Northern drill centre.

Fines content also increased significantly with depth in both 2000 and 2004. Finer particles are expected to move down-slope. This depth effect may obscure drilling effects and attenuation with distance, since depth is greater at more remote stations to the northeast of the development. For example, the highest fines values observed in 2004 occurred approximately 300 m from the Southern drill centre, but high fines values also occurred more than 20 km away, at the Northeast Reference station (the deepest station).

In general, fines content were approximately 0.5% higher in 2004 than in 2000. This increase occurred at almost every station. Since preliminary results indicate that similar increases could have occurred since 2000 at Terra Nova, the possibility of methodological discrepancies between years can not be discounted.

Sulphur, as a component of barium sulphate used in WBMs, should also be elevated where these muds are used, but background levels are high (20,000 mg/kg). Overall, any effects of drilling on fines content and sulphur did not markedly elevate values above background.

Concentrations of frequently detected metals other than barium also decreased with distance from the Southern drill centre in 2004 but not in 2000. This is assumed to be a natural change in spatial distributions. Concentrations decreased between 2000 and 2004 at more remote stations. Overall, metal concentrations were low and variance over space and time limited. The changes in spatial distribution between 2000 and 2004 were detectable only because of the large sample size and power of the study.

In summary, there was clear evidence that concentrations of $>C_{10}$-C_{21} HCs and barium were elevated by drilling activity near the Northern and Southern drill centres, and more equivocal evidence that fines content and sulphur concentrations may also have been elevated. $>C_{10}$-C_{21} HC contamination extended to between 5 and 8 km from source. Barium contamination extended to approximately 2 km from source. Any contamination from fines and sulphur was limited to within 1 km from source.

7.1.2 Biological Effects

In 2004, as in 2000, no sediment samples were toxic to the amphipods and luminescent bacteria used in standard toxicity tests.

In situ benthic invertebrate communities at White Rose were dominated by polychaetes, which accounted for approximately 75% of the organisms collected, and bivalves, which accounted for

approximately 17% of the organisms collected in both 2000 and 2004. There were some differences in sub-dominant families between 2000 and 2004. Specifically, Maldanidae (Polychaeta) and Stenoithidae (Amphipoda) were much more abundant and occurred more frequently in 2004 than in 2000, whereas the reverse was true for Cirratulidae (Polychaeta). Tanaidacea were more abundant in 2004 than in 2000. Cirratulidae were much more abundant in 2000 than in 2004, and Carditidae were relatively abundant in 2000 but were not collected in 2004.

Depth was the major correlate with invertebrate communities in both 2000 and 2004. Both diversity and bivalve abundance relative to polychaete abundance increased with depth (over a depth range of 120 to 140 m). Polychaetes were even more dominant at Terra Nova, which is located at shallower depths (90 to100 m) (Petro-Canada 2003). Depth "effects" on benthic invertebrates observed at White Rose are unlikely to be direct effects of depth related to, e.g., variation in pressure, light or temperature, which is probably minimal. Instead, depth was probably a surrogate or correlate of some other factor(s) poorly measured or unmeasured by the standard suite of sediment physical and chemical characteristics measured in the White Rose EEM and other sediment quality studies.

Sediment gravel and fines content had limited effects on invertebrate communities at White Rose. Gravel content is probably the most important natural factor affecting invertebrate communities at Terra Nova, where gravel content is higher and more variable (Petro-Canada 2003). The limited variance in gravel content removed one potential confounding factor in the White Rose EEM program.

Some community variables were significantly positively correlated with barium and sulphur concentrations in 2004. This appears to be a natural effect because the correlations extended to other metals in both 2000 and 2004. At low concentrations, some metals (e.g., zinc) are essential elements rather than toxicants, and the higher concentrations at White Rose observed in 2000 and 2004 may have mild stimulatory effects on invertebrate communities.

Overall, there was little evidence of drilling effects on benthic community variables. However, total abundance, and the relative abundance of amphipods, may have been affected by drilling. Both variables increased with distance from the Southern drill centre in 2004 but not in 2000. Total abundance also increased with distance from the Northern drill centre in 2004, but this relationship is likely natural since it was present in 2000. In 2004, the relative abundance of amphipods was also significantly (and negatively) correlated with concentrations of $>C_{10}$-C_{21} HCs.

The apparent zone of effects on total abundance and the relative abundance of amphipods extended to approximately 2 km from the Southern drill centre. However, for both variables, distance effects were mostly a function of the absence of high values, and not the occurrence of unusually low values, near the Southern drill centre. At stations greater than 2 km from the Southern drill centre, both high and low values occurred.

A number of studies carried out in the North Sea describe benthic invertebrate responses to 1 to 3 km from platforms (Olsgård and Gray 1995; Daan et al. 1994; Kingston 1992; Gray et al. 1990). In general, benthic invertebrate responses in the North Sea extend to a little less than the zone of chemical contamination. Tait et al. (2004) and Daan et al. (1996) show that this general conclusion may hold true for SBMs as well as oil-based muds, and benthic invertebrate responses extending over most of the zone of chemical contamination were also noted in the Gulf of Mexico (Montagna and Harper 1996).

With respect to HC contamination, Candler (1995) reports that values in excess of 100 mg/kg are required before benthic communities are affected; and Kingston (1992) notes that a decrease in diversity can be expected when HCs in sediments reach 50 to 60 mg/kg. HC levels at White Rose are low compared to these levels (22 mg/kg within 1 km of drill centres). However, Kingston (1992) also notes that certain sensitive species could be affected at concentrations of less than 10 mg/kg and many authors have noted that amphipods (as well as some bivalves and echinoderms) tend to be particularly sensitive to contaminants (Peterson et al. 1996; Daan et al. 1994). Therefore, the spatial extent of the potential benthic invertebrate response observed at White Rose appears to be generally consistent with the literature on effects of contamination from offshore oil development in the North Sea and in the Gulf of Mexico.

Additional sampling, as part of the scheduled 2005 EEM program, is required at White Rose to determine if noted benthic invertebrate responses truly resulted from project activity, or if they simply represent year-to-year variability in community composition. At Terra Nova, for instance, decreases in bivalve abundance near drill centres in 2001 were no longer apparent in 2002. If results seen at White Rose are project effects, then spatial differences in total abundance and the relative abundance of amphipods are not likely due to direct acute toxicity, since no toxicity was observed in laboratory tests. Rather, community effects could be due to indirect effects or to chronic toxicity involving longer exposure time.

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 2005). The Interim Sediment Quality Guidelines (ISQG) are Threshold Effects Levels (TEL) below which biological effects would not be expected to occur. The Probable Effects Levels (PEL) are levels above which effects are likely to occur. No PAH were detected at EQLs of 0.05 mg/kg, and those EQLs were lower than PELs for all PAHs (Table 7-2). However, EQLs were higher than ISQG for acenaphthene, acenaphthylene, anthracene, dibenz(*a,h*)anthracene, fluorene 2-methylnaphthalene and naphthalene. Maximum concentrations of cadmium, chromium, copper, lead and zinc were well below ISQG. EQLs for arsenic and mercury, which were not detected in any sample, were below ISQG. Again, most

metals are probably essential elements rather than toxicants at the concentrations observed at White Rose.

Table 7-2 Comparison of Measured Concentrations of PAHs and Metals to Canadian Sediment Quality Guidelines

Variable	EQL (mg/kg)	Maximum Value (2004)	ISQG (mg/kg)	PEL (mg/kg)
Acenaphthene	0.05	< EQL	0.00671	0.0889
Acenaphthylene	0.05	< EQL	0.00587	0.128
Anthracene	0.05	< EQL	0.0469	0.245
Benz(*a*)anthracene	0.05	<EQL	0.0748	0.693
Benzo(*a*)pyrene	0.05	<EQL	0.088	0.763
Chrysene	0.05	<EQL	0.108	0.846
Dibenz(*a,h*)anthracene	0.05	< EQL	0.00622	0.135
Fluoranthene	0.05	<EQL	0.113	1.494
Fluorene	0.05	< EQL	0.0212	0.144
2-Methylnaphthalene	0.05	<EQL	0.0202	0.201
Naphthalene	0.05	< EQL	0.0346	0.391
Phenanthrene	0.05	<EQL	0.0867	0.544
Pyrene	0.05	<EQL	0.153	1.398
Arsenic	2	<EQL	7.24	41.6
Cadmium	0.05	0.08	0.7	4.2
Chromium	2	7	52.3	160
Copper	1	3	18.7	108
Lead	0.5	4.0	30.2	112
Mercury	0.01	<EQL	0.13	0.7
Zinc	5	9	124	271

Notes: - Source – CCME (2005)
- CCME guidelines are not available for other variables measured at White Rose

7.2 Commercial Fish Component

7.2.1 Biological Characteristics

Overall, analysis of crab and plaice Biological Characteristics (morphometric and life history characteristics) revealed that characteristics of fish and shellfish collected in the Study Area were similar to those of animals collected in the Reference Areas. In general, there was more variability among Reference Areas, than between the Study Area and the four Reference Areas. Molting frequency (% recent molt) was highest (86%) in Reference Area 2 (the shallowest of the Areas) and lowest (3%) in Reference Area 4 (the deepest of Areas). Molting frequency was similar for Reference Areas 1 and 3, and the Study Area (50%, 42% and 40%, respectively).

7.2.2 Body Burden

With some minor exceptions, metals body burdens in crab and plaice did not differ between the Study Area and the Reference Areas, or between 2004 samples and baseline samples collected in earlier years. There was no evidence of project effects, and the 2004 data, which are more extensive than earlier data, could be treated as baseline for future EEM programs.

Differences in metals body burdens among the Reference Areas were greater than differences between the Study Area and Reference Areas, although these differences were only occasionally significant. In the absence of project effects, one would expect the four widely separated Reference Areas to differ more from each other than from the central Study Area. Overall, and for both crab and plaice, body burdens were similar over both space and time, and the Reference Areas are suitable for future EEM programs.

HCs were not detected in edible tissue (crab claws, plaice fillets) in 2004 or 2000.

In 2004, but not 2000, compounds in the $>C_{10}$-C_{21} and $>C_{21}$-C_{32} range were detected in plaice liver tissue from the Study and Reference Areas. However, PSC Maxxam Analytics (J. McDonald, pers. comm.) reports that these compounds were fatty acids rather than HCs originating from SBMs, fuel or lubricating oils.

7.2.3 Taste Tests

There was no difference in taste between Study and Reference Area plaice.

Panelists were able to distinguish between Study and Reference Area crab and preferred Reference Area crab. The interpretation of these results is not straightforward because there are strong indications that crab samples were not in good condition when tested. The Marine Institute (L. Bonnell, pers. comm.) noted that many crab were soft-shelled (or recent molts as described in Section 6), which is to be expected given that crab were collected in July. Crab had considerable blackening around the shoulder area on arrival at the Institute due to freezing raw crab rather than cooking and freezing which quickly retards enzyme spoilage. Crab were then left to thaw at 2 ºC for 24 hours which, given that crab live at temperatures near 0 ºC, would have considerably increased enzyme spoilage (J. Kiceniuk, pers. comm.). Given crab condition, triangle tests and hedonic scaling tests likely should be regarded as unreliable.

In spite of the above, comments from panelists did not indicate the presence of taint in crab. The term “taint” describes the presence or perception of an abnormal or foreign odour or taste in food (Botta 1994). The only comments relating to uncharacteristic odours or taste reported by the panelists were those related to bitterness or odour. Bitter taste is not associated with petroleum but is associated with the causal organism of ‘bitter crab disease’. Bitter crab disease is known to be

common in crab from the entire area (Dawe 2001) and is therefore not a consideration as a potential "taint" in relation to petroleum development. There were only two panelists who commented on odour, one of them correctly identified the Study Area sample but neither of them considered the odour as uncharacteristic (J. Kiceniuk, pers. comm.).

Improvement of crab sampling, storage and handling methodologies is required. Crab legs should be either cooked at sea, cooled and frozen or they should be boiled without thawing. 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.

Differences in molting frequency between shallower and deeper Reference Areas (Reference Area 2 and 4) and remaining sampling Areas has the potential to confound taste test results. If these differences persist, then consideration should be given to dropping those Reference Areas for taste tests.

7.2.4 Fish Health Indicators

7.2.4.1 Mixed Function Oxygenase

Equal numbers of immature and spent females were collected in 2004. Since maturity stage can 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 activity was analyzed separately in immature and spent female plaice from the different areas. There were no significant differences in MFO enzyme activities in spent females or immature females, as well as in males (all maturity stages pooled), among Reference Areas or between the Study and Reference Areas.

7.2.4.2 Pathology

Gross anatomy was assessed visually in all fish during the necropsies for any external or internal abnormalities or parasites. One fish from the Study Area exhibited a tumour on the head kidney and three fish from the Reference Areas displayed gill achromasia (pale gill filaments) which were confirmed by microscopy examination to be X-cell lesions.

One case of nuclear pleomorphism associated with megalocytic hepatosis and one case of oesinophilic foci were observed in the Study Area. A variety of other lesions (basophilic and clear cell foci, carcinoma, cholangioma, cholangiofibrosis, and hydropic vacuolation) associated with chemical toxicity in field and laboratory studies were not detected in either Area. Liver tissues of some fish contained myxosporean parasites but no differences among Reference Areas or between Study and Reference Areas were found. Moreover, as noted in previous years, the presence of parasites did not appear to result in any other pathological changes in hepatic tissues. The "patchy

distribution" of hepatocellular vacuolation observed in fish from all Areas was not associated with degenerative changes. Such hepatocellular vacuolation is likely linked to gonadal maturation stage (Timashova 1981; Bodammer and Murchelano 1990; Couillard et al. 1997). It is also of interest to note the presence of a cluster of X-cells in the liver of one fish.

The observations on parasitism and X-cells 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 generally absent or found only at a very low incidence in the general area.

With respect to studies on gill microstructures, a slightly higher percentage of basal hyperplasia, putatively of a background nature, was noted in the Study Area. However, microstructural changes which could be more pathological, such as epithelial lifting, extensive gill oedema, telangiectasis and lamellar fusion, were absent or found at very low frequencies in all Areas.

The presence of gill achromasia and X-cell lesions in seven plaice from the Reference Areas and one plaice from the Study Area is also 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; Mellergaard and Lang 1999; McVicar et al. 1987). Desser 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 indices, it is of interest to note from an EEM perspective, the absence or very low incidence of gill lesions associated with chemical toxicity.

Overall, the results obtained on external and internal abnormalities, MFO enzymes in liver 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 four Reference Areas. The low inter-site variability found with the various early warning bio-indicators is noted in this regard.

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 7-3) were set up to guide interpretation of results. As noted in Section 1, the "null" hypotheses (H_o) always state that no effects will be observed, even if effects have been predicted as part of the EIS.

Table 7-3 Monitoring Hypotheses

Sediment Component
H_0: There will be no change in SQT variables with distance or direction from project discharge sources over time.
Commercial Fish Component
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 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 2004 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 the Northern and Southern drill centres, and more equivocal evidence that fines content and sulphur concentrations may also have been elevated. Elevated concentrations of $>C_{10}$-C_{21} HCs and barium at White Rose were similar 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). The model predicts that HCs could be dispersed to 9 km from source(s), with most concentrations less than 100 mg/kg, and concentrations of more than 1,000 mg/kg restricted to within 300 m of drill centres. In 2004, $>C_{10}$-C_{21} HC contamination extended to between 5 and 8 km from source and the maximum concentration (275 mg/kg) was noted 300 m from the Southern drill centre. Barium contamination extended to approximately 2 km from source. Any contamination from fines and sulphur was limited to within 1 km from source.

Directional effects were noted for both $>C_{10}$-C_{21} HCs and barium in 2004, with dispersion primarily to the southeast. This is consistent with current records at White Rose for 2003 and 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.

No project-effects were noted for most benthic community variables, but total abundance and the relative abundance of amphipods may have been affected by drilling. In 2004, total abundance and the relative abundance of amphipods increased with increasing distance from the Southern drill centre and these relationships were not observed in 2000. The relative abundance of amphipods was also significantly (and negatively) correlated with concentrations of $>C_{10}$-C_{21} HCs. For both variables, distance effects were mostly a function of the absence of high values, and not the occurrence of unusually low values, near the Southern drill centre.

The apparent zone of effects on benthic invertebrates extended to 2 km, beyond the 500-m zone of effects predicted in the White Rose EIS. Nevertheless, the spatial extent of the potential benthic invertebrate response appears to be generally consistent with the recent literature on effects of contamination from offshore oil developments (see discussion in Section 7.1.2 above).

Sediment contamination and possible 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 crab and plaice morphometric and life history characteristics, were similar between White Rose and more distant Reference Areas.

7.4 Summary of Other Relevant Findings

Depth, or most likely some correlate with depth, was the major natural factor affecting invertebrate communities at White Rose in both 2000 and 2004.

There were some differences in sub-dominant families between the 2000 and 2004. Specifically, Maldanidae (Polychaeta) and Stenoithidae (Amphipoda) were much more abundant and occurred more frequently in 2004 than in 2000, whereas the reverse was true for Cirratulidae (Polychaeta). Tanaidacea were more abundant in 2004 than in 2000. Cirratulidae were much more abundant in 2000 than in 2004, and Carditidae were relatively abundant in 2000 but were not collected in 2004.

Fines content was higher in 2004 than in 2000 at almost every station.

7.5 Considerations for Future EEM Programs

7.5.1 Program Elements

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.

Changes in fines levels should be examined closely in the future.

7.5.2 Sampling and Laboratory Methodologies

Given high geopositional accuracy for sediment sampling, boxcores should be offset to avoid sampling the same area twice within or across years.

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.

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 4ºC in the dark, rather than frozen.

Testing for consistency of results when/if different taxonomists are used for benthic invertebrate identification should continue.

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.

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.

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.

7.5.3 Study Design

Adding near-field stations at approximately 300 m from the Northern, Southern and Central drill centres in 2004 increased the ability of the White Rose EEM program to detect alterations from drilling. Values of sediment physical and chemical characteristics, especially drilling mud indicators, were elevated and often extreme at the near-field stations around the Northern and especially Southern drill centres. Baseline data were not required to conclude that physical and chemical alterations had occurred there. Those stations should continue to be sampled.

The Reference stations added in 2004 varied widely in depth, which created some complications for regression analyses. More generally, natural differences in sediment characteristics in such widely

separated Reference stations are likely to be greater than natural differences among stations in the centre of the development. However, Reference sediment quality values were generally not extreme relative to values at intermediate stations. The Reference stations also provide information on natural background values and variance and are essential for the assessment of more mobile fish and shellfish.

Carry-over effects, or persistent differences among stations over time unrelated to depth and distances from drill centres, were smaller and less significant in the White Rose EEM program than in the Terra Nova EEM program. Therefore, Repeated Measures (RM) approaches and re-sampling the same stations over time may not be as effective in the White Rose EEM program as suggested in the EEM design document (Husky Energy 2004). There are still considerable statistical and other advantages to RM and re-sampling (Green 1993), and the general approach was certainly powerful enough to detect project and natural effects. At the same time, *all* stations do not necessarily have to be re-sampled every year, and stations can be added or deleted to adapt to changing circumstances over time. The approach adopted in 2004 is appropriate. A core set of 37 stations sampled in 2000 was re-sampled, but new stations were added near the drill centres and other stations were deleted.

Overall, there is no reason to substantially alter the study design for the Sediment Component of the program. The design provides adequate power to detect both project and natural effects. However, 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. Baseline data for these potential drill centres have now been collected and sampling at these centres should resume if drill centres become active. If sampling resumes around these drill centres, then any redundant station located in the immediate vicinity of drill centres should be deleted and a station at 300 m from each centre should be added (as was done for the Northern, Southern and Central drill centres once exact positions for these centres became known). Stations to be excluded from the 2005 sampling program are: NN1, NN2, NN3, NN4, NN5, SS1, SS2, SS3, SS4, SS5, and SS6.

With respect to the Commercial Fish Component, the 2004 data are more extensive than earlier data. Because no project effects were detected, body burden data could be treated as baseline for future EEM programs. 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.

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