

**ENVIRONMENTAL MONITORING PLAN  
(Biological Programs)**

**Prepared For:**

**Executive Flight Centre Fuel Services Ltd.**

**Prepared By:**



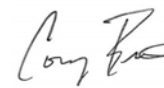
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### APPENDIX

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## 1.0 INTRODUCTION

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At the request Executive Flight Centre Fuel Services Ltd. (EFC), the Environment & Water business unit of SNC-Lavalin Inc. (SNC-Lavalin) has prepared the following Biological Monitoring Programs that will comprise a portion of the overall Environmental Monitoring Plan (EMP) for the Lemon Creek Jet-A1 fuel spill incident that occurred on July 26, 2013.

The scope of work proposed for the EMP has been developed in consideration of the results of the final Lemon Creek Spill Response Environmental Impact Assessment (EIA), which has been completed by SNC-Lavalin, on behalf of EFC, and as required by the BC Ministry of Environment (MoE). The EIA assessed and evaluated the impact(s) of the spill on physical (water, sediment) and biological (trophic levels, fisheries, terrestrial wildlife) resources. The proposed biological monitoring program herein has been identified and developed based on currently available information and SNC-Lavalin's current understanding of the conditions in the area (pre- and post-spill) as well as available research on the environmental fate of the Jet fuel/kerosene category, (e.g., American Petroleum Institute [API], 2010). Of note, although not entirely certain, we do not anticipate that chronic (prolonged) impacts to biological resources will be prevalent given the lines of evidence from the water/sediment results and our understanding on the environmental fate of the product.

The results of the water and sediment monitoring programs (programs initiated the week of August 23, 2013) will be utilized by the biological monitoring programs (where applicable) as well as human health and ecological risk assessment (HHERA) components, if appropriate. For example, in the HHERA, contaminants measured in excess of the standards/guidelines/benchmarks outlined in this document will be carried forward as chemicals of potential concern (COPCs) for evaluation in a HHERA. In addition, the results of the biological monitoring (i.e., benthic invertebrates and fisheries resources) will be considered as lines of evidence in the ERA (if required).

### 1.1 Purpose of EMP

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The key objective of the overall EMP is to ensure that the potential short-term, moderate, and longer-term (prolonged) effects to human and environmental health are effectively assessed, mitigated if necessary, and monitored for recovery. As the plan is carried out, each program will review its short-, medium-, and long-term objectives.

The EMP may also be updated, as needed, based on the outcome of each proposed, field sampling event. SCAT records and reporting (when available) have been utilized to coordinate the currently proposed focused monitoring locations and will add value on the identification of any potential new

monitoring locations. The EMP is proposed to be adaptive and continually improved/refined as results are received and evaluated.

## 1.2 Requirement for HHERA

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The biological monitoring proposed herein has been developed with consideration of the EIA, but could also provide important information for the assessment of human health and ecological risks (i.e., will inform the HHERA if required). It is noted that the EIA will assess the overall impact of the spill on the ecosystem and community, while the HHERA will specifically consider the potential for the residual contamination associated with the spill to adversely impact human health and the environment.

As indicated, the HHERA will be conducted to assess the residual contamination associated with the spill. As such, the scope of the HHERA is highly dependent on the results of the water and sediment EMP. Data collected to date indicates that current concentrations of parameters associated with the Jet-A1 Fuel are less than the laboratory detection limit in surface water and groundwater (from select domestic and agricultural wells), and that concentrations of fuel associated parameters are less than the applicable provincial standards in sediment, as well as in soil from the spill site. Based on the current data set, the residual contamination meets the applicable regulatory standards and thus, there is no need to conduct a HHERA.

As outlined in the water and sediment EMP (plan previously submitted), further investigation and sampling has been conducted (i.e., week of August 23) or is proposed to investigate the potential for: 1) pockets of residual contamination in sediments along the banks of Lemon Creek and/or the Slocan River, 2) the potential for residual contamination/fuel present in bank sediments to become solubilized when water levels rise to the levels at the time of the spill, and, 3) for dissolved phase contamination to be mobilized towards nearby wells by the pumping of the wells. Following the completion of the proposed water and sediment sampling events, the data will be reviewed to determine if concentrations in excess of the applicable standards are present, and therefore, whether or not an HHERA is required.

In addition, the results of the fish tissue and shoreline vegetation assessment (proposed below) will be reviewed to determine the requirement for the assessment of human health and ecological risks associated with the consumption of fish/impacted vegetation from the system and/or risks to these resources. If polycyclic aromatic hydrocarbons (PAHs) / alkylated PAHs are measured in the tissues, a HHERA evaluating these receptors/pathways will be conducted.

As indicated, the requirement for an HHERA will be reviewed as water, sediment, and fish tissue results are received. The scope of the HHERA will be highly dependent on the data for these various media.

## 2.0 OBJECTIVES

The objectives of Biological Monitoring Programs of the EMP are to:

- 1) assess and document the distribution and concentrations of residual contaminants associated with the spill in fish to determine the extent of the impacts and to ensure all potentially impacted human and ecological receptors (endpoints) are identified and evaluated; and
- 2) assess and monitor the effect of the spill on key biological (primarily aquatic) indicators (endpoints) as well as recovery of those indicators that were (and/or potentially) impacted;

### 2.1 Identification of Recovery Endpoints

An endpoint is a measured response of a receptor to a stressor and can be measured in a toxicity test or field assessment.

Recovery endpoints were established through consultation with provincial agencies (MoE, MFLNRO), First Nation (Canadian Columbia River Inter-tribal Fisheries Commission), as well as local stakeholders (Slocan River Streamkeepers) and fisheries professionals (Mirkwood Ecological Consultants) with vast knowledge of the Slocan River system. The endpoints recognize pertinent findings from emergency response phase, those biological features that are highly valued in the region, and the importance for effectively monitoring the status and recovery of aquatic health post-spill.

Recovery endpoints for the riverine environment are presented in Table 2.1 along with the criteria used for selecting the endpoints.

**Table 2.1: Recovery Endpoints for the Riverine (Aquatic) Environment**

Component	Endpoint	Rationale and criteria for selection
Fish Consumption	PAH levels in mountain whitefish from the Slocan River and Brilliant Dam are similar to background levels in Little Slocan River, or with a PAH profile dissimilar to the released product. There is no fish tainting.	Mountain whitefish has previously been used as an indicator species in the Columbia system. Given their importance to the local recreational fishery, this species was chosen as the target species. A broad spectrum of science has been established that all teleost fish have a well developed capacity to metabolize PAHs. Because of the efficient metabolism, there is very low potential for PAHs to accumulate in muscle, thus low potential for transfer of PAHs up the food chain to human and wildlife consumers. Further, fish tainting has been included on the chance that consumers raise concerns on how fish taste.

Table 2.1 (Cont'd): Recovery Endpoints for the Biological (Aquatic) Environment

Component	Endpoint	Rationale and criteria for selection
Fisheries Resources	Population estimates of mountain whitefish in the Slocan River near Lemon Creek were not significantly adversely impacted. Fish abundance and community structure in braided (side-channels) of the Slocan River near Lemon Creek are similar to mortality counts in the same area and are temporally consistent and stable.	The majority of deceased fish specimens collected during the emergency response were located within the off-channel braided watercourses just downstream of the Slocan River-Lemon Creek confluence. Given the highest species count of deceased fish were of mountain whitefish, it is anticipated that they are the most abundant species inhabiting Slocan River off-channel habitat and were most impacted as a result of the spill.
	Rainbow trout populations in the Slocan River were not adversely impacted.	Rainbow trout populations of the Slocan River have been in serious decline, but more recent studies have suggested population recovery. Previous studies of Slocan River index sites found the highest numbers of rainbow trout near Lemon Creek, about half as many fish at Winlaw, about one quarter as many fish, less at Crescent Valley, and lowest numbers of fish at Passmore and Slocan Park (Oliver, 1999). Population monitoring has been ongoing including post-spill; however, insufficient funds has prevented the analysis of current and historic data.
	Bull trout migration and spawning in the Lemon Creek system have not been adversely impacted.	Bull trout are a blue-listed (species of concern) fish species in BC. No bull trout mortalities were collected during seven days of post-spill salvages. However, the system is believed to be a highly productive spawning area and likely provides habitat for other life stages.
	Fish abundance and community structure in Lemon Creek are similar to reference (non-impacted) sites.	Lemon Creek is one of, if not, the most diverse and productive watercourses for fish species in the Slocan River system. It is home to several important fish species including bull trout (Blue-listed), rainbow trout, mountain whitefish, sculpin spp including shorthead sculpin (SARA listed), and umatilla dace (also SARA listed). The lower 4 kilometres of Lemon Creek were the most highly impacted from the spill based on SCAT data/results and deceased fish specimens collected during salvage.
Lower Trophic Level Dynamics	Similar density, distribution, diversity, and biomass of benthic invertebrates in affected and reference areas as well as regional data (where feasible).	Benthic invertebrate community structure represents an important ecosystem health indicator and well as an indication of aquatic recovery post- impact event.

Aquatic impacts and recovery as a result from, and post-clean up of, this type of fuel spill has previously been studied by Guiney et al. (1987) and the American Petroleum Institute – Petroleum HPV Testing Group (API, 2010).

The HPV Chemical Test Program of the American Petroleum Institute (API, 2010) included acute toxicity endpoints for freshwater fish, freshwater invertebrate, and freshwater alga for jet fuel/kerosene category. The substances in the Jet fuel/kerosene were found to produce a similar range of toxicity for each of the three trophic levels (API, 2010) and there is sufficient data on the ecotoxicity of jet fuel and kerosenes to demonstrate moderate acute toxicity to aquatic organisms. This is predicted because the majority of constituents in kerosenes are neutral organic hydrocarbons that act in a common mode of action termed “non-polar narcosis”, which is brought about by disruption of biological membrane function (van Wezel and Opperhuizen, 1995). Thus, it is anticipated that any chronic toxicity effects or impacts to species, populations, or communities of these organisms to be low.

Further, there are currently only minor (if any) detectable concentrations of contaminants of concern, and no exceedences in surface water and sediment collected from Lemon Creek and the Slocan River, which indicates that these two media are likely no longer adversely influenced (SNC-Lavalin, 2013a). However, seasonal monitoring of surface water and sediment will occur (SNC-Lavalin, 2013a) to verify this finding; as such, the proposed biological monitoring programs have also taken into account current conditions as well as analytical data collected during the initial spill response phase.

The above product information, results from the emergency response and EIA, extensive literature, and local knowledge/expertise was compiled and evaluated prior to selecting the monitoring components and associated indicators including rationale for their inclusion as well as defining the scope of each program.

The following sections outline the proposed scopes to meet the above objectives and carry out each program.



## 3.0 BIOLOGICAL MONITORING PROGRAMS

### 3.1 Fisheries Resources

There are three key objectives that will comprise the Fisheries Resource Monitoring Plan. These include: (1) the evaluation of fish health and concerns around human (and wildlife) health, (2) assessment of key fisheries indicators (e.g., species-level, population-level, community-level); and (3) detection and identification of the recovery process based on results from (1) and (2).

The populations and community of fish that inhabit both Lemon Creek and the Slocan River system were impacted as a result of the spill, based on the evidence of fish mortalities collected during the emergency response. However, the magnitude of the impact is currently unknown. It is known that jet fuel/kerosene is moderately acutely toxic to fish (as well as to benthic invertebrates and algae; API, 2010). Data from the provincial veterinary lab on causation of death as part of the emergency response phase (SNC-Lavalin, 2013) confirmed acute lethal toxicity to fish (and benthic) species in both Lemon Creek and Slocan River. SCAT observations (Polaris, 2013) along with field observations by professional biologists during the fish salvage program also observed substantial mortality of benthic invertebrates.

Based on evaluation of emergency phase field observations and SCAT findings (Polaris, 2013) and the Emergency Response Management Workbook (Quantum Murray, 2013) key focal areas for short- and longer-term monitoring have been identified on Lemon Creek as well as the mainstem and braided segments of the Slocan River downstream of Lemon Creek confluence to Appledale (the braided reach). The lowermost 4 kilometres of Lemon Creek received the full effects of the fuel spill event, with no significant off-channel refuge habitat downstream of the spill. Thus monitoring programs have been developed to evaluate the Lemon Creek mainstem and key (select) tributaries. The Slocan River monitoring programs will focus on both select braided reaches the mainstem. Highest concentrations of accumulated hydrocarbon product were found in the braided reach just downstream of the Slocan River-Lemon Creek confluence, which is where the majority of fish mortalities in the Slocan River were observed. The area provides a diversity of quality habitat, with an abundance of instream and overhead riparian cover.

Priority fish species for consideration were rainbow trout (*Oncorhynchus mykiss*), mountain whitefish (*Prosopium williamsoni*), and bull trout (*Salvelinus confluentus*). These species have been partially studied by local streamkeeper groups (primarily rainbow trout in the Slocan River) providing information on previous monitoring methodology and background data, which has been considered in the identification and development of biological programs, proposed methodology, and analyses. Species significant to

the local recreational fishery and abundant in the area of impact include mountain whitefish and northern pikeminnow (*Ptychocheilus oregonensis*). Mountain whitefish, in particular, were prominent in the recovery of fish mortalities and have been included as a measure of site recovery post-impact. There are also three fish species-at-risk known to potentially inhabit the impacted area, including white sturgeon (*Acipenser transmontanus*), shorthead sculpin (*Cottus confusus*), and Umatilla dace (*Rhinichthys umatilla*). The believed low abundance of these species pre-spill reduces the suitability of these species as indicators of site recovery. A number of deceased sculpin and dace specimens were collected as part of the salvage program; these specimens are currently undergoing formal species ID to confirm whether either species were impacted by the spill. We have not included a monitoring program for these species at this time, but should species ID of deceased specimens confirm their presence, then further evaluation and discussions will occur with local regulators to see whether a monitoring program is feasible.

Timing of each proposed monitoring program considers either key life history timing and strategies of the agreed upon targeted species, or repeating previous assessments for before-impact data comparison. Further, the development of the biological monitoring programs solicited, and has taken into consideration, input and comments provided by local First Nations (Canadian Columbia River Inter-tribal Fishery Commission), stakeholders (i.e., Slocan Streamkeepers), and regulators (MFLNRO, MoE). It is SNC-Lavalin's intent to implement the proposed biological monitoring programs through a collaborative effort with local stakeholders to take advantage of local knowledge and expertise.

### 3.1.1 Fish Tissue Assessment Program

During the spill response program a limited number of fish were collected from high exposure areas and were submitted for laboratory analysis of PAHs and alkylated PAHs. The fish have been filleted by the laboratory, with a composite sample of tissues from the three fish analyzed, and the remaining portions, including organs, archived for future analysis if required. The results of the analysis have not been received to date.

An additional fish tissue sampling program is proposed to further assess the potential for the jet fuel released to Lemon Creek to be present in the tissues of fish, and thus, represent a potential risk to both human and wildlife consumers. The proposed fish tissue program will include the collection of the following:

- 1) Five (5) live "catchable" (> 20cm) mountain whitefish from within the Slocan River, near the Lemon Creek confluence;
- 2) Five (5) live "catchable" (> 20cm) mountain whitefish from within Brilliant Reservoir; and

- 3) Three (3) live “catchable” (> 20 cm) mountain whitefish from the Little Slokan River (reference site) where there was high likelihood that the collections were not influenced by the spill.

The sample size of 10 (five impact samples from Slokan River near Lemon Creek confluence and five impact samples from the Brilliant Dam reservoir) has been shown to be adequate to distinguish “exposed” from reference sites in other studies (Swanson, 1985; USEPA, 1993).

The mountain whitefish has previously been used as an indicator species in the Columbia system (McPhail & Troffe 1998). Given their importance to the local recreational fishery, we have chosen mountain whitefish as the target species for this program. Lemon Creek is closed to fishing year round, thus specimens from this system have not been included in the assessment. However, deceased sub-adult mountain whitefish specimens collected from Lemon Creek during the emergency response phase fish salvage program have been sent for fish tissue analysis, which will provide informative toxicity information from specimens inhabiting the highest impact zone. Recreational fishing regulations for Slokan River limit trout and char to catch and release from September 1 to March 31. However, there is a daily quota of 15 mountain whitefish in Slokan River from September 1 to March 31. Thus, whitefish consumption safety is of primary concern in the areas closest to the spill, hence our focus on this species. Further, given the conservation importance/concern of rainbow trout in the Slokan River system, we have deliberately excluded this species in the tissue assessment and feel that results from mountain whitefish will apply to other species.

It is noted that based on the surface water chemistry results collected during the spill response phase, parameters associated with the Jet-A1 fuel have been less than the laboratory detection limits since August 3, 2013. Of the PCOCs associated with the Jet Fuel-A1, PAHs have the potential to partition to fish tissues (or be taken up by fish). The metabolism of PAHs by teleost fish is well documented in the scientific literature, and has been discussed by Canadian agencies in their derivation of surface water guidelines for PAHs (BC MoE, 1993<sup>1</sup>; CCME, 1999<sup>2</sup>). The US Department of Commerce, National Oceanic and Atmospheric Administration presented the findings of a scientific review evaluating the metabolism of PAHs by teleost fish. Their summary indicates that there is broad foundation of science established documenting the metabolism of PAHs by teleost fish. They also indicate that because of this efficient metabolism, there is very low potential for PAHs to accumulate in the muscle tissue, and therefore, low potential transfer up the food chain.

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<sup>1</sup> BC MoE Ambient Water Quality Criteria for Polycyclic Aromatic Hydrocarbons (PAHs). February 1993. Available at <http://www.env.gov.bc.ca/wat/wq/BCguidelines/pahs/index.html#TopOfPage>.

<sup>2</sup> CCME Canadian Water Quality Guidelines for the Protection of Aquatic Life, Polycyclic Aromatic Hydrocarbons. 1999. Available at <http://ceqg-rcqe.ccme.ca/>.

Given the low potential for bioaccumulation, the short exposure duration for fish in Lemon Creek/the Slocan River (i.e., given the rapid decrease in concentrations to levels below the laboratory detection limits) and the time that has passed since detectable concentrations were measured, it is our opinion that the number of fish tissue samples proposed is sufficient to assess potential fish consumption exposures/associated risks (for both human and wildlife consumers).

#### 3.1.1.1 Fish Tainting

SNC-Lavalin understands that the BC MoE had recommended the consideration of fish tainting (i.e., potential impacts of the spill on the taste of fish from the area). One of the project laboratories has indicated that they can conduct objective analyses to evaluate the potential for fish tainting, however, there are no standards or guidelines for the results to be compared to. Despite this, we propose to proceed with the analyses, in the event that complaints from the public regarding the taste of fish caught from the area are made; the results could provide context to the complaints (i.e., are they related to perceived risk, is the tainting associated with the spill?). It is noted that in a memo dated August 23, 2013, the MoE suggested that the Environmental Monitoring Plan should include consideration of fish tainting. On this basis, we propose proceeding with the analysis.

#### ***Specimen Sampling Methodology and Timing***

We propose to deploy a quick gill net set in Brilliant reservoir to capture the desired number of specimens (mountain whitefish). For collection of specimens in the Slocan River system, we will apply either a gill net (based on site conditions) or angle in index sites where mountain whitefish are known to be abundant. All other specimens collected by use of either gill net or angling will be documented, but released unharmed.

Given the mountain whitefish fishery opening within the Slocan River commences early September, we propose to target sample collection in mid- to late-September. We anticipate it will take 1-2 field days to collect the fish samples from the three locations.

#### ***Lab Analysis***

The results of the fish tissue samples collected to date, as well as data currently being generated by the provincial veterinary lab regarding causation of death, and toxicity screening data on collected (deceased) fish specimens during the emergency response will be reviewed in the evaluation of analytical screening requirements for live captured specimens.

Whole specimens will be submitted to the project laboratory. Prior to analysis, the laboratory will fillet the fish, with the need for composite samples determined based on the size of the captured specimens. Tissue samples will be analyzed for PAHs, including alkylated PAHs. Organs will be archived, with the requirement for future analysis determined based on the tissue chemistry results or other observations indicative of potential exposures.

If tissue chemistry results and/or other observations are indicative of exposures, consideration will be given to analysis of livers from the specimens for PAHs/alkylated PAHs, and/or analysis of ethoxyresorufin-O-deethylase (EROD) activity<sup>3</sup>.

### **3.1.2 Slocan River Mainstream and Off-Channel Fish Monitoring Program**

#### **3.1.2.1 Slocan River Off-Channel Mark-Recapture Monitoring**

This section summarizes the proposed mark-recapture study in off-channel (braided) habitats of the Slocan River.

The purpose of the mark-recapture program is to characterize post-spill salmonid populations in select off-channel habitats on the Slocan River. The majority of deceased fish specimens collected during the emergency response were located within the off-channel braided watercourses just downstream of the Slocan River-Lemon Creek confluence. Given the highest species count of deceased fish were of mountain whitefish, it is anticipated that they are the most abundant species inhabiting Slocan River off-channel habitat, and will be the most abundant species captured, thus the mark-recapture program will primarily target this species. All other species of fish captured as part of this program will be documented with the aim to contribute to data sets for those species that inhabit the Slocan River. Given there is extremely limited historic information on mountain whitefish abundance and population estimates for the Slocan River, the data generated from this program will primarily be compared to impact estimates. However, we do propose that this program continue in 2014 to allow for some limited temporal comparison between years to evaluate the scale of impact to mountain whitefish and monitor for potential recovery.

This proposed mark-recapture program has been developed based on the review of procedures and analyses described in Brittain et al. (1997) Rosenberger & Dunham (2005), Triton (2007), and Golder (2010).

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<sup>3</sup> EROD is a catalytic measurement of cytochrome P4501A induction, and is used as a biomarker for PAH exposure in fish.

## ***Biophysical and Biological Data Collection***

### Water Quality

During the mark-recapture field component, in-situ water quality parameters including conductivity, dissolved oxygen, temperature, and pH will be recorded at both the upstream and downstream end of each “closed” sampling site.

### Channel Characteristics

Each sample site will be measured and marked every 25 metres with flagging tape. Wetted width and mean depth will be recorded at each 25 m “transect”, as will the presence/absence of habitat features, with changes in substrate composition noted. Substrate categories will be defined using the Udden-Wentworth grain-size scale following Buffington & Montgomery (1999).

### Site Characteristics

Between 3-5 “closed” sites will be selected within each of the identified braided channels (index sites).

### Fish Sampling & Marking

Fish abundance estimates from electrofishing capture data are most often generated using mark-recapture models (Thompson et al. 1998). We propose to collect fish specimens by using a backpack electrofisher and adopting methods applied in Rosenberger & Dunham (2005), which generally includes selecting sites that are 100 metres in length that will be blocked off at the upstream and downstream ends of the sites (secured to streambed). To evaluate potential bias from escapement (a violation of the closed population assumption), we will equip our sites with double block nets at both the upstream and downstream ends, following Peterson et al. (2004). Either a 3- or 4-pass removal technique will be applied.

Fish will be marked by using a BMX2000 POW'R-Ject BioPhotonic (or alternative) Marking system (NEWEST Technologies). *BMX2000BioPhotonicMarking* is a “state-of-the-art” technique in tagging of a wide range of macro-organisms. It is the least invasive, externally visible, non-destructive marking technology available for wildlife research, assessment and management. This highly robust *spectral marking* technique for identifying and monitoring diverse species in the wild has also been approved by the US Food and Drug Administration (USFDA) for industrial/commercial applications.

Created for placement beneath translucent skins of animals and plants or surface mounting on highly pigmented, non-translucent or rigid integuments, *BioPhotonicTags* (“BPTs”) are made with the highest quality fluorescent microspheres. These particles are mixed in a proprietary formulation that assures bio-compatibility and high retention as well as ease and safety of administration even under difficult field conditions.

The use of this marking technique has been proven successful in other monitoring studies of juvenile fish species (e.g., Triton 2007, Scott Hinsch, UBC pers. comm.).

### ***Spatial and Temporal Extent***

#### Site Locations

As mentioned above, the following off-channel habitats along the Slocan River will be evaluated as part of the mark-recapture program. They include:

- ◆ The upper braided reach of Slocan River to 1.5 km downstream of the Lemon Creek confluence; and
- ◆ The braided channel downstream from the Winlaw bridge.

The index site downstream of Winlaw had the closest comparable channel morphology to the upper impacted braided areas near the Lemon Creek confluence. Further, both these sites may compliment data from the upper two reaches of the proposed mainstem Slocan River rainbow trout snorkel surveys. Juvenile rainbow trout presence in the associated side channels can be inferred from past snorkel survey data from the main channel of these selected index sites.

#### Sampling Frequency

Each watercourse will be sampled twice, first to mark fish and second to enumerate marked and unmarked fish. Sampling to “mark” fish will be conducted in late-September 2013. Biophysical assessments (described above) will be conducted immediately following the marking component of the program. The enumeration of marked/unmarked fish will occur in early October, no more than 4 days (96 hours) after fish have been marked. These dates coincide with juvenile mountain whitefish sampling conducted in the lower Columbia River (Golder 2010). Further, electrofishing can alter fish behaviour for up to 24 hours (Mesa & Schreck 1989), and can be exacerbated by handling and tagging fish. Thus, an inadequate recovery period may result in an unequal capture probability between marked and unmarked fish, violating a mark-recapture model assumption. To minimize for potential bias from changes in fish behaviour after marking and handling (violation of the equal capture probability assumption of the mark-recapture model), we will vary the time between marking fish and the first removal (recapture) pass between a range of 24 hours and 48 hours.

Given that there is no pre-spill baseline data to compare results with, we propose to conduct the same program again in late September 2014. This will provide opportunity to compare data over two seasons and aid in better understanding the magnitude of impact to the mountain whitefish population(s).

## Data Compilation & Analyses

### Biological Data

All captured fish will be enumerated and identified to species. Fish will be anaesthetized using clove oil for collection of biological data. Fork length will be measured to the nearest millimetre using a measuring board and weights to the nearest 0.01 g using a balance.

Only those mountain whitefish and/or rainbow trout that are >60 mm will be marked. Specimens <60 mm will be measured, weighed, and documented but will remain unmarked.

A common practice in fisheries surveys is to record the lengths and weights of captured fish for analysis and to calculate the condition of fish. Condition is a measure of the well-being, fatness and/or gonad development of a fish (Blackwell et al. 2000), which provides insight into the environmental conditions available to fish (e.g., habitat, prey, predators). “Fat” fish usually indicate favorable environmental conditions, whereas “thin” fish usually indicate unfavorable environmental conditions where fish are more susceptible to negative changes in their environment (e.g., more susceptible to pollutants).

Over the years, several methods have been developed to estimate the condition of fish. One of the most popular approaches is Fulton K, which is defined as (Anderson and Neumann, 1996):

$$K = \frac{W}{L^3} 100,000 \text{ Equation 1}$$

Where W is the weight in grams, L is the length in millimetres and 100,000 is a constant used for scaling purposes (Blackwell et al. 2000).

This measure has been used for decades to describe fish condition. It is however limited by the assumption of isometric growth (i.e., equal growth patterns among different size classes), which is rarely the case (Blackwell et al. 2000). Relative weight ( $W_r$ ) is another method that has been introduced in the last decade and is usually preferred because it corrects the bias incurred with Fulton K. It requires a species-specific equation ( $W_s$ ), which is available for mountain whitefish (Rogers et al. 1996). The equation for mountain whitefish is:

$$\log_{10} W_s = -5.086 + 3.036 \log_{10} TL \text{ Equation 2}$$

where  $W_s$  is the weight in grams and TL is the total length in millimetres. It should be noted that Rogers et al. (1996) found no evident systematic length bias in relative weight values calculated with this equation and regional trends in condition were not observed.



### Population Estimates

Sampling data will be used to derive population estimates for mountain whitefish (and potentially rainbow trout) as well as generate descriptive statistics for other species where appropriate. These will include (but are not limited to):

- ◆ Number of fish marked versus the number recovered for mountain whitefish;
- ◆ Densities of fish in all sampling areas (e.g., fish/m<sup>2</sup>);
- ◆ Mean and standard deviation of length, length:weight, and length:frequency data; and,
- ◆ Habitat preferences in the surveyed areas, where sufficient data exists to show preference.

Population estimates of juvenile mountain whitefish will be calculated using a modified Schnabel Method (Ricker 1975) and the sequential Bayes Algorithm (Gazey & Staley 1986). This analysis is consistent to what has been performed on juvenile mountain whitefish in the Columbia River (e.g., Golder 2010). The following assumptions will be applied for these population estimates:

- ◆ The population is closed (i.e., the population remains the same size over the period of the program);
- ◆ The probability of capturing a marked individual at a given time is equal to the proportion of marked members in the population at that time;
- ◆ Fish do not lose their marks during the study period, and
- ◆ All marks are reported on recovery.

#### 3.1.2.2 Slocan River Mainstem Rainbow Trout Population Analysis

The Slocan Rivers Streamkeepers have been conducting long term monitoring of a variety of environmental and biological variables in the Slocan River to serve as a baseline from which to monitor ecological change. Spawning productivity of rainbow trout (*Oncorhynchus mykiss*) was identified as a reasonable measurement of ecosystem health and, therefore, has been used valuable biological variable that has been monitored over several years. The intent of the monitoring surveys was to collect data for comparison from year to year at the same location (relative abundance) versus determining total productivity through out the river.

This years (2013) rainbow trout field (snorkel) surveys were recently completed for the Slocan River. However, through discussions with Peter Corbett (Mirkwood Ecological Consultants) who has been leading the rainbow trout surveys on the Slocan River for the last several years, funding to analyze the data collected (including comparing data across years) has been extremely limited. Thus, results are

lacking over the last several years to fully understand the status of rainbow trout populations in the Slocan River system. Given the timing of the recent field surveys took place post-spill, the remaining gap to be filled is analyzing data. SNC-Lavalin will work with Mirkwood Ecological Consultants to ensure appropriate data analyses are completed. Results will be reported through Mirkwood Ecological Consultants to SNC-Lavalin. In turn, SNC-Lavalin will ensure the results of the report are included as part of the scheduled interim and final reporting as described in Appendix I.

### **3.1.3 Lemon Creek – Mainstem and Tributary Assessment Program**

A priority fish species for consideration in Lemon Creek is bull trout (*Salvelinus confluentus*). No bull trout mortalities were collected during seven days of post-impact salvages. However, the system is a known spawning location and may provide habitat for other life stages. Also of concern are mountain whitefish and rainbow trout, with whitefish consisting of the highest portion of direct fish mortalities recovered from Lemon Creek.

Information in literature is limited with respect to bull trout numbers, population dynamics, utilization, or timing within the Lemon Creek system. Previous information suggests the mouth of Lemon Creek could be utilized by juvenile bull trout in the fall and winter. Bull trout were not recorded in recent Slocan River snorkel surveys downstream of Lemon Creek, completed by the Slocan River Streamkeepers. Local observation indicates the spawning bull trout are possibly an adfluvial population from Slocan Lake that delay migration to Lemon Creek until water temperatures in Slocan River decrease to approximately 15°C, which tends to occur in late September through October (Peter Corbett, pers. comm.). This timing also coincides with low flow periods and decreasing water temperatures in Lemon Creek. Bull trout are anticipated to spawn in accessible habitat within Lemon Creek and its tributaries. Bull trout have very specific habitat requirements for spawning and rearing. Bull trout depend upon water temperatures below 9°C to begin spawning (McPhail and Baxter 1996). Long incubation periods make spawning site selection especially important for bull trout. Optimum temperatures for incubation are reported to be between 2°C and 4°C (Goetz, 1989 from Berge and Mavros, 2001). Emergence of bull trout fry typically occurs in the late spring, and these fish may stay in their natal stream for up to 4 years.

In a 1996 forest stream inventory, rainbow trout were abundant in the lower reaches of Lemon Creek, while bull trout were more prevalent in upper reaches (Corbett, 1996). Spawning and rearing habitat were observed throughout the Lemon Creek mainstem.

### 3.1.3.1 Bull Trout Redd Survey

Spawning survey data are important for determining relative abundance and distribution in bull trout populations. The proposed sampling plan should provide a snapshot of spawning activity immediately post-impact, while many years of data would be needed for determining bull trout population trends (Sausen, 2013).

Objectives of the bull trout spawning surveys include:

- ◆ Verify bull trout migration and spawning in Lemon Creek has proceeded in the first post-impact season;
- ◆ Determine bull trout timing of spawning;
- ◆ Locate and map bull trout spawning areas;
- ◆ Determine and compare the spatial distribution of redds;
- ◆ Determine if there are bull trout redd locations in areas of Lemon Creek downstream of the spill;
- ◆ Determine redd characteristics;
- ◆ Collect spawning density data;
- ◆ Determine and compare bull trout and whitefish spawning overlap (if feasible); and
- ◆ Assess for further fish mortalities within and upstream of the impacted zone of Lemon Creek.

Office desktop tasks prior to the redd assessments will include a detailed review of literature and local knowledge of bull trout spawning. The reach break data available for Lemon Creek from past stream classification studies will be reviewed. Mainstem and tributary reaches will be measured and categorized for suitability of assessment as index areas. One additional separate reference location on the Little Slocan system will be selected. The use of a known spawning location in an unimpacted system would verify the sampling method is successful in detecting redds should there be negative results from the Lemon Creek sites.

Fall redd assessments will be completed on foot with accepted field reconnaissance methods. Previous data on expected bull trout numbers or spawning timing in Lemon Creek and its tributaries is unknown at this time. Different locations with bull trout of differing life history types spawn at a range of timing from September through December. Based on limited information on timing of spawning, three rounds of assessment will occur, once in the first week of October, again in the third week of October, and a final assessment in the first week of November. A high flow event may necessitate postponing surveys due to unsafe conditions until flows recede.

Six (6) surveyors will be partnered in crews of two and assigned index areas to sample. The fairly large area anticipated to be accessible by bull trout for spawning will require assessments over multiple days. We intend to focus field efforts on two tributaries (Monument Creek and Holmsen Creek) as well as a stretch of Lemon Creek mainstem (near river kilometre 18) what are believed to represent the primary spawning areas for bull trout in the Lemon Creek system. We will also deploy hoop nets at around kilometre 12 on Lemon Creek to confirm upstream and downstream migration of spawning bull trout and/or kelts (post spawn condition). We intend to deploy hoop nets daily and check nets at the end of each day after completing redd surveys.

The survey teams will be comprised of SNC-Lavalin biologists as well as partners from Mirkwood Ecological Consultants. Each survey team will work concurrently in different assigned areas. Surveyors will walk in a zigzag pattern across wide channel reaches of the river and along the bank when the entire wetted channel can be viewed from shore. When a redd is located in the field the site is marked by hanging a piece of flagging on the adjacent bank.

Data Collection will occur on datasheets printed on rite-in-the-rain paper. Redd measurements data recorded for each redd includes the following: width of the “head” of each redd, the width of the “pit” at center of the redd, the width of the “tailspill”, and total length of the redd. Data recorded during the bull trout spawning surveys included; 1) date of survey, 2) stream location, 3) size of redds, 4) visibility of redds, 5) number of redds, and 6) approximate number and sizes of bull trout and other species (particularly mountain whitefish) observed during surveys (Barnett and Paige, unpublished). Reach break locations will be documented as they relate to the index sites. The location of each redd or cluster of redds will be georeferenced using a handheld GPS. Each redd location observed in the first sample session will be marked with flagging tape for relocation during subsequent surveys. All stream flagging will be removed during the final surveys.

Bull trout redds are counted in a manner to avoid overcounting. Redds will be categorized as “definite” or “probable” to account for test/false redds. The redds will be assessed for loose eggs on the surface and by gently tapping the tailspill with a wading stick to determine if it is “soft” or not as compared to gravel next to the redd (Barnett and Paige, unpublished). Survey team will not dig in the site to disturb eggs. A site that has been disturbed by a spawning bull trout will move easily and not produce large amounts of sediment when tapped.

Bull trout are known to cluster redds in preferred locations with potential superimposition or chaining. Signs that redd superimposition occurred include visibly new gravels deposited on top of an old tailspill, or seeing fish actively working at the site. Bull trout may also use the pit of a previous redd as an egg pocket and then bury eggs leading to a chain of redds in an upstream direction. The tailspill of the

upstream redd fills the pit of the downstream redd leading to chaining, with each site a separate redd. In order to call a new redd, evidence such as new gravels in the tailspill material (not colored by algae, completely free of sediment), fish active on the new redd site, clear headwall cut that hasn't yet collapsed or sloughed away (with time, these definite boundaries fade) should be assessed. If it cannot clearly be determined that this be the case, the redd will be recorded as a single long site (Barnett and Paige, unpublished).

Mountain whitefish spawning season is expected to, at least, partially overlap with the bull trout spawning season. Unlike bull trout, mountain whitefish are broadcast spawners that release their eggs over areas of coarse gravel and cobble; they do not build redds. General information will be collected and will attempt to estimate the number of fish in a particular school if feasible.

Also recorded will be an estimate of the depth of water where the whitefish are located and a description of the macrohabitat type at the site (e.g., pool, riffle, glide).

It should be noted that bull trout spawning surveys are also being performed in tributaries around Slocan Lake at approximately the same time as proposed for Lemon Creek surveys. It is our understanding that Mountain Water Research will be conducting these surveys (Jeff Burrows, pers. Comm.). We have connected with Mountain Water Research to ensure that our proposed methodologies are consistent between the programs to ensure appropriate comparability of data but without compromising the objective(s) of the bull trout redd survey proposed herein.

### 3.1.3.2 Fish Abundance & Community Recovery Monitoring

Assessment of recovery of fish abundance and community composition in the impacted reach will be completed by a combination of open and closed site sampling (e.g., Triton, 2006). Closed sites are selected locations where seine nets can be used to block fish accessing or leaving the survey area during the survey. Assessment sampling will take a control-impact approach, with multiple reference sites providing the "control" conditions anticipated to have occurred pre-impact.

The proposed fish-sampling plan will include a combination of G-type minnow trapping and electrofishing. Where feasible, pole seining may be conducted to supplement the proposed program; however, it is anticipated the large substrate size will limit the ability to conduct effective seining.

These studies were initially proposed to be completed on three occasions. However, given bull trout spawning migration to commence soon/is occurring in Lemon Creek, will prevent the use of electrofishing as a mode of sample collection. Thus, the proposed late September 2013 sampling will be delayed until August 2014. The late August 2014 field sampling would target a range of bar/bank edge habitats and assess juvenile distribution while the April 2014 field sampling, to evaluate overwintering

habitat use and possibly salmonid fry post-emergence habitat selection (Triton, 2008). Given the results from the April and late August 2014 sampling, it is possible that additional monitoring will be required repeating in 2015 to further assess fish community recovery in the impacted portions of Lemon Creek. The spring timing is intended to occur prior to freshet when visibility and safety concerns may limit mainstem sampling. Earlier timing would be limited by site access, particularly due to snowpack at higher locations in the drainage.

Fish sampling is proposed to be conducted by a three person sampling crew. A three person crew will be required in order to transport and effectively deploy sampling gear within difficult to access reaches, as well as collect physical habitat characteristics data. For efficiency, while two people are electrofishing, the third person will complete note-taking, setting and checking traps, etc. Sample sites will follow the index area approach determined for the bull trout redd assessments.

The remote location with poor communications ability will restrict sampling to daylight hours to address access and safety concerns. Use of volunteers to assist with execution of this (and other) proposed programs will be considered but it is more logical that local biologists (e.g., Mirkwood Ecological Consultants) with previous knowledge of the study area will be used to execute the field program. Therefore, it is expected all field work will be conducted by qualified professionals.

### ***Minnow Trapping***

Minnow traps will be set during each sampling episode. Traps will be baited with salmon roe and will be set at a density of 10 traps/100 m in each sampled index area (Triton, 2008). The traps will be baited with salmon roe and set overnight for approximately 24 hours. Trap locations will be marked in the field and georeferenced using hand held GPS units. Other data recorded at each trapping location will include hydraulic morphology unit type (riffle, pool, glide), trap depth, proximity of nearby cover features and cover type.

### ***Electrofishing***

Electrofishing will be conducted during each sampling event. Sampling sites will be selected to represent various habitat types within each site and will be sampled by multiple pass removal to maximize opportunities for identifying species composition and fish densities in surveyed areas. Each electrofishing site will be measured to nearest metre, geo-referenced, and marked with flagging tape at the upstream and downstream end for replication on future surveys. Accessibility for closed site electrofishing will be field determined. We have targeted late August 2014 for sampling to avoid possible encounters with migrating spawning bull trout.

### **Biological Data Collection**

Captured fish will be anaesthetized, identified to species and enumerated by size classes. Captured fish will be measured for length (to the nearest mm) and weight (1/100 gm). Data forms will be developed to ensure consistency of data collection. Captured fish will be allowed to recover in flow-through buckets and will be released back into the stream near the vicinity of capture. Voucher specimens will only be collected from any inadvertent mortalities during sampling and handling, or if mortalities are encountered not resultant of the sampling (Triton, 2008).

Fish sampling data will be compiled to provide the following information:

- ◆ Relative abundance of fish (by species) in the sampling areas (expressed as fish/m<sup>2</sup>) and densities of fish in different habitat units (pool, run, glide);
- ◆ Diversity including number of species and % catch by species;
- ◆ Catch Per Unit Effort (CPUE) and % catch by species;
- ◆ Average fork length (mm);
- ◆ Approximate age to length, length to weight and length to frequency data; and
- ◆ Habitat selection preferences in surveyed areas by species.

## **3.2 Lower Trophic Level Dynamics**

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### **3.2.1 Slocan River and Lemon Creek Benthic Invertebrate Program**

#### **3.2.1.1 Benthic Invertebrate Recovery Monitoring**

Benthic invertebrates are small animals that live on or in the bottom sediments of waterbodies and are an important source of food for fish. In most benthic community assessments, the primary objective is to determine the identity, abundance, and distribution of the species that are present (USEPA 1992a; 1994). Because most benthic macroinvertebrates are relatively sedentary and are closely associated with the sedimentary environment, they tend to be sensitive to both short-term and long-term changes in habitat, sediment, and water quality conditions (Davis and Lathrop 1992). Therefore, data on the density, distribution, diversity, and biomass of these species provide important information on the health status of aquatic ecosystems. As such, benthic invertebrate community structure represents an important ecosystem health indicator.

Particular insect taxa are considered to be more sensitive to pollution than others and, therefore, these taxa have been used in various bioassessment studies to develop biotic indices (Woodiwiss, 1964; Hilsenhoff, 1988; Kerans and Karr, 1994). These pollution sensitive taxa include *Ephemeroptera* (mayflies), *Plecoptera* (stoneflies) and *Trichoptera* (caddisflies), collectively referred to as EPT taxa. Streams with higher relative abundance of pollution-sensitive taxa typically receive a higher “score” in these biotic indices (e.g., B-IBI, Hilsenhoff, 1988) than streams with lower levels of these taxa. Biological indices have been applied to invertebrate taxa, which have been exposed to pollution exposure over months and years, at varying levels of toxicity.

The benthic invertebrate community in Lemon Creek and the Slocan River will be assessed and monitored as individual systems and will be sampled post-spill to provide an estimate of the:

- ◆ Measurable impacts, if any, resulting from exposure to Jet Fuel A-1 (based on Shell MSDS); and
- ◆ Recovery in the weeks and months after the spill.

As mentioned above, it is known that jet fuel/kerosene is moderately acutely toxic to benthic invertebrates, API 2010). SCAT observations (Polaris 2013) along with field observations by professional biologists during the fish salvage program also observed substantial mortality of benthic invertebrates.

### **Approach**

“Recovery” in this context will be defined as returning to pre-spill condition(s). In the absence of benthos data from the hours immediately prior to the spill, this study will apply the following comparisons for evaluating recovery:

- ◆ A Before-After-Control Impact (BACI) study design using both Lemon Creek (where possible) and Slocan River pre-spill benthos data (Before), re-sampling of these sites after the spill (After) and including sites upstream (Control) and downstream (Impact) of the spill;
- ◆ Regional data (i.e., Reference Condition Approach [RCA] using Canadian Aquatic Biomonitoring Network [CABIN] Reference Site for Columbia River Basin);
- ◆ A Lemon Creek control site upstream of the spill site, as well as a Slocan River control site upstream of the Lemon Creek confluence. These sites will be sampled on the same dates as downstream sampling sites; and,
- ◆ Intra-river sample comparisons over a minimum of three kick net sampling periods after the spill (e.g., Analysis of Similarities, Similarity Percentages, Analysis of Variance).

Comparing samples from locations exposed to Jet Fuel A-1 with samples from locations within the same watercourse without exposure (i.e., upstream control) is a common approach in lotic studies.



Historic benthos data pre-spill are available for both Slocan River and, to a much lesser extent, Lemon Creek. There is one site on Lemon Creek that has been previously been sampled in the late 1990's using a Hess Sampler (Jennifer Yeow, personal communication). This data has been requested so it can be considered as pre-spill baseline. If data is provided, we intend to apply both the CABIN protocol and Hess sampling within Lemon Creek. The CABIN protocol will be the only method to be applied to data collection on the Slocan River.

Estimating the spill-related changes in the benthic invertebrate community presents several challenges including:

- ◆ Distinguishing between natural variation and the stress of the spill on the benthic community; and
- ◆ Inherent natural variability among and between sites based on site-specific factors (e.g., substrate type).

Based on the data collected as part of this proposed monitoring program, we intend to address some key questions. These include:

- ◆ What are the differences in community composition, density, biomass and relative abundance?
- ◆ How much of this difference can be attributed to expected year to year variation?
- ◆ How do the inter-site similarities within the benthos communities of the affected areas?
- ◆ Compare with the similarities between the control site and affected communities across?
- ◆ The sampling dates?

### ***Proposed Methodology***

#### Literature Review

A review of existing literature from benthic invertebrate studies conducted previously on Lemon Creek and Slocan River will be conducted. These sources will include (but are not limited to):

- ◆ 1:20,000 terrain resource information mapping (TRIM) for the area or similar (i.e., satellite imagery);
- ◆ BC MoE Ecological Reports Catalogue (EcoCat) website (BC MoE 2013);
- ◆ Winlaw Watershed Committee;
- ◆ BC Ministry of Environment – Nelson office;

- ◆ Slocan Streamkeepers Society;
- ◆ First Nations; and
- ◆ Other consultant reports.

### Field Program

The BC provincial government has specific guidelines on how to effectively sample benthic invertebrate in freshwater environments (see Beatty et al. 2006). However, the Canadian Aquatic Biomonitoring Network (CABIN) through Environment Canada will be the primary driver of the benthos monitoring program. CABIN is an aquatic biological monitoring program for assessing the health of freshwater ecosystems in Canada. A set of national protocols are used for field collection, laboratory work, and analysis of biological monitoring data. Individuals who collect data under CABIN must be certified through Environment Canada's program training. SNC-Lavalin has professional biologist's (RPBio's) with this certification and will lead in the development of the detailed field design, site selection, data collection, and analyses.

The following are those guidelines/standards that will be referenced and modified (where applicable) to supplement the CABIN protocols. They include:

- ◆ BC Field Sampling Manual (Clark, 2003);
- ◆ Guidelines for Monitoring Benthos in Freshwater Environments (Gibbons et al., 1993); and
- ◆ Guidelines for Sampling Benthic Invertebrates in British Columbia Streams (Beatty et al., 2006).

As benthic invertebrate diversity and abundance is typically most abundant during the late summer/early fall months, sampling will be conducted in early October (2013) to coincide with early fall conditions. Overall, we propose three sampling events. These include:

- ◆ Early October 2013 to coincide closely with historical sampling;
- ◆ March 2014 to coincide with Beatty et al. (2006); and
- ◆ Early October 2014.

The 2013 benthos field program will be conducted concurrently with other biological monitoring components (e.g., fish sampling), where possible, to ensure efficiencies thus reduce costs.

Benthic invertebrate samples are proposed to be collected from the following two main watercourses:

- ◆ Lemon Creek:
  - 2 reference sites upstream of the spill site. These sites will serve as control sites to which benthic invertebrate data collected in downstream impacted reaches will be compared; and,
  - 4 sites located between the spill site and the Slocan River-Lemon Creek confluence.
- ◆ Slocan River:
  - 1 reference sites upstream of the confluence with Lemon Creek and downstream of Slocan Lake;
  - 1 reference site in the Little Slocan River and 1 reference site in Bonanza Creek; and
  - 6 sites between the Slocan River-Lemon Creek confluence and the Winlaw Bridge.

Of note, the proposed number of sample sites selected is consistent with that applied on a similar spill monitoring program in southwest BC (Triton, 2008). Further, there are specific locations within the Slocan River system (e.g., Slocan River, South Slocan River, Bonanza Creek etc.) that have pre-spill benthos data collected (through CABIN), which have preliminarily been evaluated and included as sample (reference) locations in this program.

The specific location of each site will be chosen during the field program and will be based on habitat conditions of each sampling area (i.e., Lemon Creek and Slocan River). For example, sites will be selected in an area of each watercourse based on a combination of BC provincial guidelines (Beatty et al., 2006) and the CABIN: Wadeable Streams Field Manual (Environment Canada, 2012).

#### Sampling Methodology

Sites will be sampled in an upstream direction to avoid potential contamination (i.e., sample collection will start at the Lemon Creek/Slocan River confluence and proceed upstream to the spill site and further upstream to the reference area). All benthic invertebrate samples will be sieved in the field and shipped in coolers with ice packs to the taxonomist.

As per CABIN protocols, one independent grab (replicate) will be sampled relatively close to each other using a Hess sampler (ONLY for the site where a Hess sampler was historically used on Lemon Creek) or a kicknet (for CABIN sites) in an area where riffle habitat is present, as per CABIN protocols. Replicate samples are recommended to be collected in order to assess sample heterogeneity and reduce natural variability (Beatty et al., 2006). Observations on the appearance of the sample (e.g., colour, odour, presence of material) will be recorded. Each sample will be sieved in the field using a 250 µm - 400 µm screen, and fixed in 10% formalin prior to shipping.

The site locations will be geo-referenced and photo-documented to allow for future samples to be collected at the same location. This will allow a spatial and temporal comparison of results.

Collection methods will follow CABIN protocols. Additional information collected during the benthic invertebrate study will include biophysical data such as stream velocity, channel width, substrate, channel morphology, in-situ water quality, and fish habitat cover. In essence, biophysical parameters required by CABIN will be collected.

#### Data Analysis

Benthic invertebrate samples will be shipped to Ruxten Environmental Consulting in Vancouver, BC and enumerated and identified to the lowest practicable level (ideally genus and species). Taxonomic analysis will be performed on each individual replicate sample. Results presented will include determining indices, through CABIN, of:

- ◆ Percent abundance – a comparison of the relative abundance of all taxa;
- ◆ **EPT** – percent and number of Ephemeroptera, Plecoptera, and Tricoptera taxa;
- ◆ Simpson’s Evenness Index – indicates the degree to which all taxa of the same level are equal in abundance;
- ◆ Shannon-Wiener Diversity – quantifies the degree of uncertainty that a specific taxon will be present; and,
- ◆ Simpson’s diversity index – the proportion of each taxon relative to the total number of invertebrates in a sample as a measure of taxa richness.

#### Quality Assurance (QA)/Quality Control (QC)

QA/QC for laboratory analyses will follow CABIN Laboratory Methods Manual (<http://www.ec.gc.ca/rcba-cabin/default.asp?lang=en&n=6DBF844E-1>). The description of QA/QC prepared by Glozier et al. (2002) will also be referenced (as appropriate). There will be three components to QA/QC; sorting efficiency, sub-sampling precision, and sub-sampling accuracy.

### **3.3 Terrestrial Wildlife Resources**

Currently, we propose to continue with performing necropsies on terrestrial wildlife specimens collected during the emergency response phase to better understand any potential linkages between the spill (and its associated compounds) and cause of death. Further, field observations will continue through the implementation of all biological and physical field programs. If any additional deceased specimens are

observed in the field, they will be identified to species, photodocumented, characterized for level of decomposition, collection location marked (GPS), appropriately stored to preserve the specimen, and be considered further for causation of death screening. Specimens collected henceforth will then be discussed with EFC and regulators prior to any further action taken.

## 4.0 REPORTING

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A proposed (tentative) schedule has been developed for initiating the biological monitoring programs (see Appendix I) and reporting. Given the number of upfront field programs proposed for early fall, data entry, analysis, and interpretation will not commence until after field work is completed. We propose to provide interim reporting of select preliminary results by January 31, 2014 (See Appendix I). It is our intent to report results back on the analytical findings with respect to fish tissues as quickly as possible once results have been received from the certified lab and have been analyzed by appropriate SNC-Lavalin technical experts. We anticipate mid-/end October 2013 results can be provided. We will also provide a second round of interim reporting by July 1, 2014. Finally, we propose to provide final reports by July 31, 2014. For those programs that are proposed to continue after July 1, 2014 (benthics mark-recapture, fish abundance & community programs) interim and final reports will follow the same schedule as described above.

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## 6.0 LIMITATIONS OF LIABILITY, SCOPE OF REPORT AND THIRD PARTY RELIANCE

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# APPENDIX I

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Proposed Schedule 2013 / 2014



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