Revised Proposal for Ecotoxicological Testing of Clontarf Bay Site

Prepared for

Coffey Environments

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Prepared by

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Introduction

This proposal will address and fulfil the Ministerial Conditions for East Clontarf Residential Subdivision No 7 Water Quality

Following is a Scope of Work that will address the requirements of subsections 7-4, 7-5 and 7-6 of the ministerial statement:

"7 Water Quality

7-4 Prior to ground-disturbing activity, the proponent shall prepare an Ecotoxological Testing Plan to monitor the benthic habitat at the wetland discharge point into Clontarf Bay to the requirements of the Minister for the Environment on advice of the Environmental Protection Authority.

7-5 The proponent shall implement the Ecotoxological Testing Plan required by condition 7-4.

7-6 The proponent shall make the Ecotoxological Testing Plan required by condition 7-4 publicly available."

Background

The proposed residential subdivision bound by Manning Road to the north, Centenary Avenue to the east, the Clontarf Aboriginal Education and Training College to the west, and Canning River (Clontarf Bay) to the south has the potential to release contaminants into the Canning River. If the ground is disturbed there is potential to release heavy metals and total petroleum hydrocarbons, thus increasing the contaminant load in the Canning River at the wetland discharge point.

A baseline study to assess the current state of the wetland discharge point has been performed using a unicellular algal growth test (Geotech Report ENV06-193) in August 2006 and a Microtox bacterial growth test (Geotech Report ECX07-2703) in March 2007. These results are shown in Table 1. Results for the microalgal growth inhibition assay showed that growth was enhanced in all concentrations of groundwater, where some constituents of the samples were acting as nutrients. Therefore, the nutrients within the samples may be ameliorating or masking the effects of any toxicants present. The Microtox results showed that sites MW4 and MW5 exhibited toxicity to the bacteria. These two sites are closest to a former landfill site so would be expected to show some toxicity. However, these two sites are furthest from the wetland discharge site and would be unlikely to impact on the wetland discharge.

Table 1 Toxicity Test Results

Site	Algal IC50 %	Microtox EC50 %
MW1	>72.4	No Sample
MW2	>72.4	No Sample
MW3	>72.4	>90.9
MW4	>72.4	42.5
MW5	>72.4	34.9
MW6	>72.4	>90.9
MW7	>72.4	>90.9
Canning River Water	>100	>90.9

Proposal for Further Testing

A range of bioassays using different species from several different trophic levels are commonly used to assess environmental samples following the ANZECC and ARMCANZ (2000) Water Quality Guidelines. Several species are used to produce a species sensitivity distribution as the sensitivities of different species varies eg. one species may be very sensitive and another may be very tolerant, as shown in the preliminary tests. The species protection trigger values are then calculated from the species distribution curve using the CSIROs BurrliOZ statistics program.

Following the protocol outlined in ANZECC and ARMCANZ (2000) 99%, 95%, 90% and 80% species protection trigger values will be calculated using the EC10 data from a minimum of five chronic bioassays (5 species from 4 different trophic levels) that will be performed on species indigenous to, or surrogate to, the receiving ecosystem in the Canning River.

As the salinity of the Canning River varies from 10 ppt (as measured in August 2006) to 41 ppt (as measured in March 2007), the benthic communities at the discharge site would not remain static throughout the year. As the salinity changes, the organisms living in or on the benthos would move to a more suitable site. Therefore, performing tests on Canning River organisms is not feasible due to the changing community structure and salinities. To overcome this problem Geotech proposes to perform a suite of bioassays using freshwater species indigenous to or surrogate to, the wetland to characterise the toxicity of the discharge. Geotech also propose to assess the toxicity of the sediment using an indigenous amphipod (*Grandiderellia* sp.).

Proposed Bioassays

Geotech proposes to use Microtox as a screening test and then as a routine monitoring test and a suite of freshwater bioassays listed in Table 2.

Microtox

The basic technology of the Microtox Test System is based upon the use of luminescent bacteria, specifically the strain *Vibrio fischeri* NRRL B-11177, to measure toxicity from environmental samples. When properly grown, luminescent bacteria produce light as a by-product of their cellular respiration. Cell respiration is fundamental to cellular metabolism and all associated life processes. Bacterial bioluminescence is tied directly to cell respiration, and any inhibition of cellular activity (toxicity) results in a decreased rate of respiration and a corresponding decrease in the rate of luminescence. The more toxic the sample, the greater the percent light loss from the test suspension of luminescent bacteria. Bacterial bioluminescence has proved to be a convenient measure of cellular metabolism and consequently, a reliable sensor for measuring the presence of toxic chemicals in aquatic samples. Strain 11177 was originally chosen for the acute and chronic tests because it displayed a high sensitivity to a broad range of chemicals.

Table 2. Freshwater Bioassays

Test	Species	Duration	End Point
Unicellular Alga	*Chlorella sp.*	72 hours	Growth
Duckweed	*Lemna sp.*	7 days	Growth
Daphnia	*Ceriodaphnia dubia*	7 Days	Reproduction
Copepod	*Macrocyclops albidus*	26 days	Reproduction
Larval Fish	Pygmy Perch	7 days	Growth

Unicellular Alga Growth

Algae are primary producers of organic matter upon which animals depend either directly or indirectly through the food chain. As such, test procedures using algae are valuable for determining the primary productivity of a water samples and for testing the toxicity of chemicals present in the water. The chronic toxicity test using marine microalgae are used to measure the effect of test materials on growth over a 72 h period. This test has the added benefit of measuring stimulatory as well as inhibitory effects. Geotech is proposing to use the common microalgal species *Chlorella protothecoides*. In Australia, the tests using *Chlorella protothecoides* have been widely used along side invertebrate toxicity tests.

Copepod Reproduction

The freshwater Copepod (*Macrocyclops albidus*) is commonly found in freshwater systems around Perth. Copepods are the most common crustacean which live in the plankton and eat unicellular algae. This bioassay exposes more than one life stage to the discharge. This reproduction bioassay using the copepod is similar to the USEPA 7 day *Ceriodaphnia*

dubia reproduction tests and the methodology is based on the USEPA Method 1002.0. This test assesses the reproductive ability of a female exposed to the toxicant from the neonate stage.

Daphnia Reproduction
As above similar to the copepod reproduction using the USEPA Method 1002.0 7 day Daphnia reproduction test.

Duckweed Growth Test
Duckweed (*Lemna* sp.) is a common plant in freshwater systems around Perth. Duckweed provides food and shelter for many freshwater organisms and is an important part of the ecosystem. This test uses the ASTM E 1415-91 (Reapproved 2004) methodology and uses growth after 7 days as the end point.

Larval Fish Growth
The test organism, the pygmy perch (*Edelia vittata*) is one of the most common and widespread freshwater fishes endemic to south-western Western Australia. It is found in rivers, streams, lakes and pools and is associated with riparian vegetation*. E. vittate* feeds on daphnia and copepods. The *E. vittata* larvae are obtained from naturally spawning commercial broodstock. Broodstock are fed a varied diet to ensure good quality larvae. The eggs are collected each morning and the quality determined. Only larvae from high quality eggs are used in the bioassays. The newly hatched larvae are grown for 7 days and the growth determined and used to calculate the EC50.

Amphipod Sediment Bioassay
Amphipods (a small “sand flea”-like crustacean) are an important component of the benthic community, proving food for birds, fish and larger invertebrates. Geotech proposes to perform a sediment bioassay using the local amphipod *Grandidierella* sp. with a sample of sediment taken from the site and compared with a control sample. This bioassay measures growth of the amphipod after 14 days exposure to the sediments.

Statistical Calculations
All statistical calculations are performed using the Tidepool Scientific ToxCalcv5. The concentration of discharge affecting 50% and 10% of the population (EC50 and EC10) will be determined by a Probit analysis or the trimmed Spearman-Karber Method. The concentration causing no observed effect (NOEC) and the lowest concentration causing an effect (LOEC) will be determined using an analysis of variance followed by Dunnetts or a non-parametric test, depending on normality of distribution.

The EC10 values of the freshwater tests will be used to calculate a protection value (99%, 95%, 90% or 80%) as determined by the DEC using the BurrliOZ statistics package as recommended by the ANZECC and ARMCANZ Water Quality Guidelines (2000).

Monitoring

In order to fulfil the Ministerial statements 7.4 Geotech will perform a suite of five bioassays on the species listed above following ground disturbance and a rain event. This will determine if toxicity has changed due to an increase in contaminants leaching from the site to obtain a "worst case scenario". The EC10 values will be used to calculate the species protection trigger values. The trigger values will be calculated using a concentration of an easily measured contaminant such as zinc or aluminium as a representative of the concentration of discharge, to be determined at the time of testing. These chemical concentrations can be monitored when the monthly Microtox tests are performed.

A routine monitoring program is then recommended which involves screening of the site on a regular basis using Microtox on a monthly basis for 12 months and a three monthly basis after completion of construction. If any changes are detected then a full investigation (a full suite of toxicity tests) and management procedures are implemented.

The results from this monitoring can be used to trigger further testing or implementation of management procedures if results are significantly above those obtained from the baseline data assessment. Sediments are long-term integrators of contaminants and need only to be monitored annually.