ENVIRONMENTAL

Nitrogen losses via denitrification after irrigating treated wastewater at Omaha Beach – technical memorandum

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Action	Name	Date
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Contents

Executive Summary

The fate of nitrogen following irrigation of treated wastewater at Jones Rd and the Golf Course is a key issue at Omaha. Whilst the wastewater is well-treated, stakeholders are concerned that nitrogen leached from the irrigation areas enter Whangateau Harbour, which could potentially lead to water quality degradation in this high quality waterbody.

Previous desktop studies done on this issue (Diffuse Sources, 2008) concluded that significant nitrogen input to the harbour was unlikely because:

(i) travel time between the irrigation sites and the harbour was of the order of 22-35 years from the golf course and 9-11 years from Jones Road,
(ii) the presence of high organic matter in the Kahikatea forest adjacent to the golf course, and peat deposits both beneath the forest and in discontinuous layers away from the Jones Road site favoured denitrification (a process whereby the oxidized forms of nitrogen – nitrite and nitrate, are reduced to nitrogen gas, which is lost to the atmosphere), and,
(iii) there is a high degree of tidal exchange, so any biologically-available nitrogen entering the harbour would have a short residence time – i.e. it would flushed from the harbour faster than algae could grow.

This study reports on field and laboratory measurements that provide initial data on point (ii) above. Denitrification enzyme activity (DEA) was measured at between 3-5 depths at 15 sites both within the irrigation areas and at other sites towards the harbour chosen to be representative of the flow path.

For denitrification to occur, there needs to be (i) an absence of oxygen, (ii) sufficient available carbon to sustain the energy requirements of denitrifying microorganisms, and (iii) the presence of nitrate. In anaerobic environments without carbon limitations, the amount of enzyme produced is proportional to the concentration of nitrate available. Redox potential (a measure of oxidation-reduction state of environment) was measured at water table depth, as a surrogate measure of whether the dissolved oxygen or nitrate was present, or whether more 'intense' reduction processes were present (which would also not favour denitrification).

The results showed distinct differences in DEA profiles at the two irrigation sites. At the Jones Road site DEA was measureable throughout the profile (1 – 2 m depth), and whilst variable, rates of at least 10 ng N/g/h were measured (up to 57 ng N/g/h). The highest rates were generally measured in the surface layers (but not always). The Jones Road sites were generally organic-rich peat and sand throughout the profile. The redox potential (Eh) at the water table of indicated aerobic conditions. At the Golf Course irrigation area, DEA was only measurable in the surface layer (0-200mm). This is not surprising as only this layer contained any organic matter, with the 'soil' quickly grading to medium textured sand below ~200 mm depth. One site was aerobic at the water table, however Eh at the other site (HA10) matched the state where oxygen should be

absent, but nitrate present. This coincided with organic staining of the sand, which might indicate a source of available carbon. Despite this, there was no measureable DEA.

In the transect away from the Jones Rd irrigation area measureable DEA was recorded at all depths at HA4 (closest to the irrigation) area, but only in the 0-200mm layers at HA5 (closest to the harbour). We measured the highest DEA of any sample (210 ng N/g/h) in the surface layer at HA5. While there was a moist amorphous plastic layer at about 200mm depth (which may indicate perched water) no groundwater table was encountered down to at least 2 m depth. Both of these sites had peat layers within the profile.

Down gradient (towards the harbour) from the golf course more variable environments were encountered. At the forest fringe (golf course side) the DEAs at site PAX and PCX were very similar to their golf course counterparts (HA6 and HA10), indicating no significant change in environmental conditions that might influence denitrification (nitrate, organic matter, oxygen status). On the far side of the forest (FEB1a and HA9) the soil was noticeable wetter and the Eh indicated anaerobic conditions in groundwater (HA9 only) together with organic staining of sand. DEA was only detected in an organic silt layer (200-300mm). At FEB1a DEA was measureable down to ~600mm depth corresponding to noticeable organic staining in the profile.

At the fringe of the salt marsh (HA12), a relatively high DEA (35 ng N/g/h) was recorded in the surface layer and there was measureable DEA down to 1 m depth. At sites further towards the harbour, lower DEA values were recorded and redox potential at the water table indicated aerobic conditions.

The combination of DEA results, Eh measurements and soil profiles indicate that s denitrification occurs within the organic peat soils at the Jones Road irrigation area itself. Even though Eh at the water table indicates aerobic conditions, the organic nature of the soils mean that it likely at anaerobic conditions would exist within the soil particles and that denitrification would occur (albeit diffusion-limited). Available carbon is key to driving the denitrification process and even though there is clearly a plentiful carbon supply it's availability is unknown. At the Golf course, conditions are clearly unsuitable for denitrification on the irrigated area, though they are very suitable for nitrification of residual ammoniacal-nitrogen in the irrigated effluent. The sites selected down gradient of the golf course probably do not reflect the flow path of leachate, particularly PAX and PCX. On the far side of the forest conditions are more suitable for denitrification but we have no certainty that it comes into contact with WWTP leachate.

We recommend that a revised scope be prepared for Phase 2 of this work, taking into account greater certainty on the flow path of leachate provided by the PDP groundwater model. The scope should include more in-depth studies at the Jones Rd site, which may be sufficient in itself to effect high N removal, sampling within the Kahikatea forest itself (rather than the fringes), and measurements of readily mineralisable carbon. There may also be merit in conducting short-term nitrification assays at the irrigation sites to provide confirmation that residual ammoniacal-N in the effluent is converted to nitrate, although this can probably be reasonably inferred.

1. Introduction

Watercare Services Ltd (Watercare) is in the process of preparing an Assessment of Environmental Effects (AEE) for renewal of their consent for operation of the Omaha Wastewater Treatment plant. The existing consent expired in May 2014, so a temporary replacement application was lodged in November 2014. A more detailed application is now being prepared, following consultation with a Consultative Group of stakeholders.

Following consultation with stakeholders, a revised investigation plan was issued, which included a workstream to better understand nitrogen removal processes in soil and wetlands between the irrigation sites and Whangateau Harbour. Streamlined Environmental were asked to undertake denitrification enzyme activity assays at sites selected for geohydrological investigations. The first stage of this work was to:

- In conjunction with the groundwater studies recommended in the workbrief, install at least 3-4 bores with an auger along at least one transect from each site (Omaha Golf Course and Jones Rd) to the Harbour.

- Undertake denitrifying enzyme activity (DEA) assays on duplicate soil and wetland cores from each of the sites and measure the increase in nitrous oxide over time as an estimate of denitrification rates. The initial set of samples will be taken over summer with as many assays carried out as possible in the irrigation areas and between these areas and the Harbour, over a 3 day period.

After email exchanges on the 4th and 5th December 2014, it was agreed to modify the brief as follows:

- Foregoing the duplicate cores and rather separating the cores by depth (the rationale being that if nitrate from the WWTP is present it may be in a band corresponding to the water table. Take 3-4 samples per single core at each hand-augered core site.

The soil/wetland sampling was carried out on the 13th and 14th January 2015 with the assays performed over the following 3-4 days.

This technical memorandum reports on the results of the assays. We intend that these results feed into:

1. Planning for the second stage of work identified in the investigation plan, and,
2. Calculations on nutrient loading to the estuary and the AEE as appropriate.

2. Methods

PDP consultants (Aslan Perwick) supplied us with the GPS coordinates for the sites used for geophysical investigations (Figure 1). In addition two further sites (PAX and PCX), which were at existing monitoring bores inside the predator fence on the DOC reserve, were added during the sampling after discussions with Nicholas Woodley. The draft bore logs prepared by PDP were used as an initial guide to select sampling depths but it soon became apparent that the depth to water table had increased significantly in the month between PDP doing their investigations and our sampling. There was, however, very good agreement between the PDP logs and our observations (with the exception of water table depth). We used the soil logs as a guide for selecting

3-5 sampling depths at each site. In general these corresponded to surface (0-200 mm), 400-600 mm, 800-1000 mm and at the water table + 200 mm. We used a hand auger to take samples. When the target depth was reached samples were placed in plastic bags, labelled, sealed and bagged again before being placed in a chilly bin for transported on ice back to the laboratory.

It was only possible to measure redox potential (Eh) in the deepest sample (at water table). Where redox potential was measured, the sample was place in a plastic bag, and the electrode (platinum) inserted into the centre of the sample. A reading was taken after the current stabilised. The electrode was calibrated against a Thermo Scientific Orion ORP standard (cat. No. 967961) where E= 220mV @ 25C and Eh= 420mV at 25C for a Normal Hydrogen Electrode. The electrode was washed with deionised water between readings and the calibration checked regularly.

At the laboratory the bagged soil samples were mixed by kneading the bags and stirring with a mixing spoon. Denitrification enzyme activity (DEA) was measured by the acetylene block technique (acetylene blocks the conversion of nitrous oxide to nitrogen during denitrification) using a modification of the method described by Cooke and White (1987). DEA provides a surrogate measure of the activity of denitrifying enzymes *in situ* (i.e., enzymes active at the time of sampling), and therefore is indicative of the historic site conditions. The rate represents not only enzyme activity, but also the environmental factors that control enzyme expression (oxygen content, C availability, and nitrate concentration). In anaerobic environments without carbon limitations, the amount of enzyme produced is proportional to the concentration of nitrate available, and the rate of N_2O production is proportional the enzyme content.

A subsample of soil was weighed and transferred to a monovette (volume 35mL, Figure 2) and a second subsample weighed for dry weight determination. The monovette containing the soil was flushed three times with oxygen-free nitrogen gas. This was followed by the addition of 0.5mL acetylene gas and 5mL of 20 mmol/L $NaNO_3$. The nitrate solution was sparged with nitrogen gas to deoxygenate the sample prior to use. The sample was shaken vigorously for 2 mins and then transferred to a rotary shaker for continuous mixing at 20˚C. A 2mL gas sample was removed at specified time intervals and transferred to 3mL vaccutainers for Gas Chromatograph-Electron Capture Detector (GC-ECD) analyses.

Gases were analysed on a Shimadzu GC-17A GC fitted with an ECD. Calibration was performed routinely using N_2O gas standards manufactured by BOC Gases (1ppm, 10ppm and 30ppm). The GC was fitted with a Porapak T pre-column (25cm x 1/8 inch dia), a Porapak Q (6 feet x 1/8 inch dia) packed column and Vico valve which enables back-flushing of water vapour from the pre-column. GC injector temperature was 55˚C, column oven temperature was 50˚C and ECD temperature was 330˚C. Oxygen-free nitrogen carrier gas was used at a flow rate of 30 mL/min. Injection loop volume was 0.2mL. Detection limit on GC was approx. 0.2 ppmv (Atmospheric levels of N_2O in air are typically 0.33 ppmv). N_2O concentration in the headspace over the 2 hours (0, 0.5h, 1h, 2h) was plotted, corrected for dry weight, N_2O dissolved in equilibrium with solution using a Bunsen adsorption coefficient at 20^o C of 0.644 (Carter and Gregorich, 2007) and expressed as µg N_2O-N /g of soil.

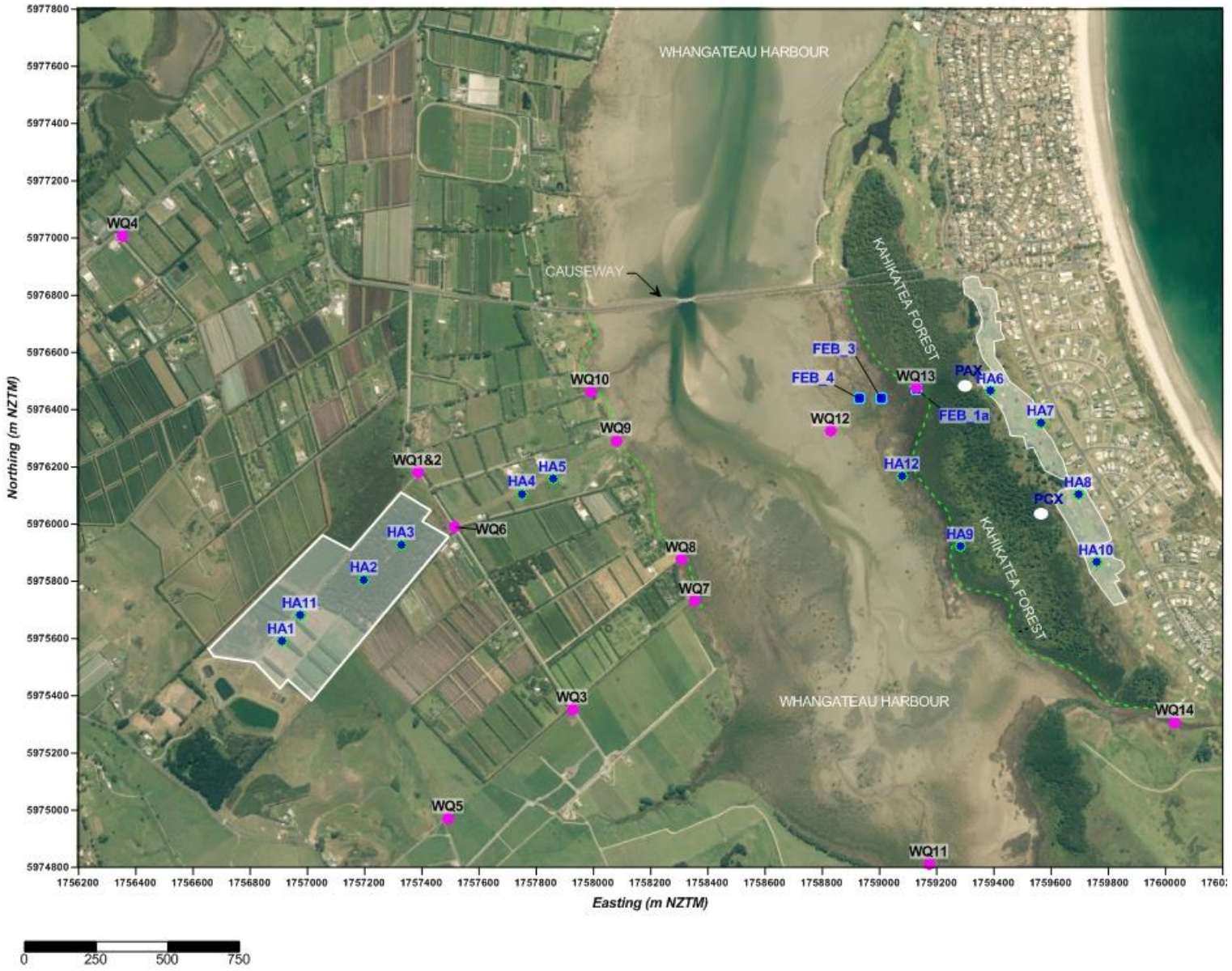


Figure 1 Soil sampling sites for DEA assays (HA 1-10 (excluding HA7 and HA8), FEB1a, 3 and 4, PAX and PCX). Note WQ (pink sites are surface water quality sampling sites and not sampled for DEA.

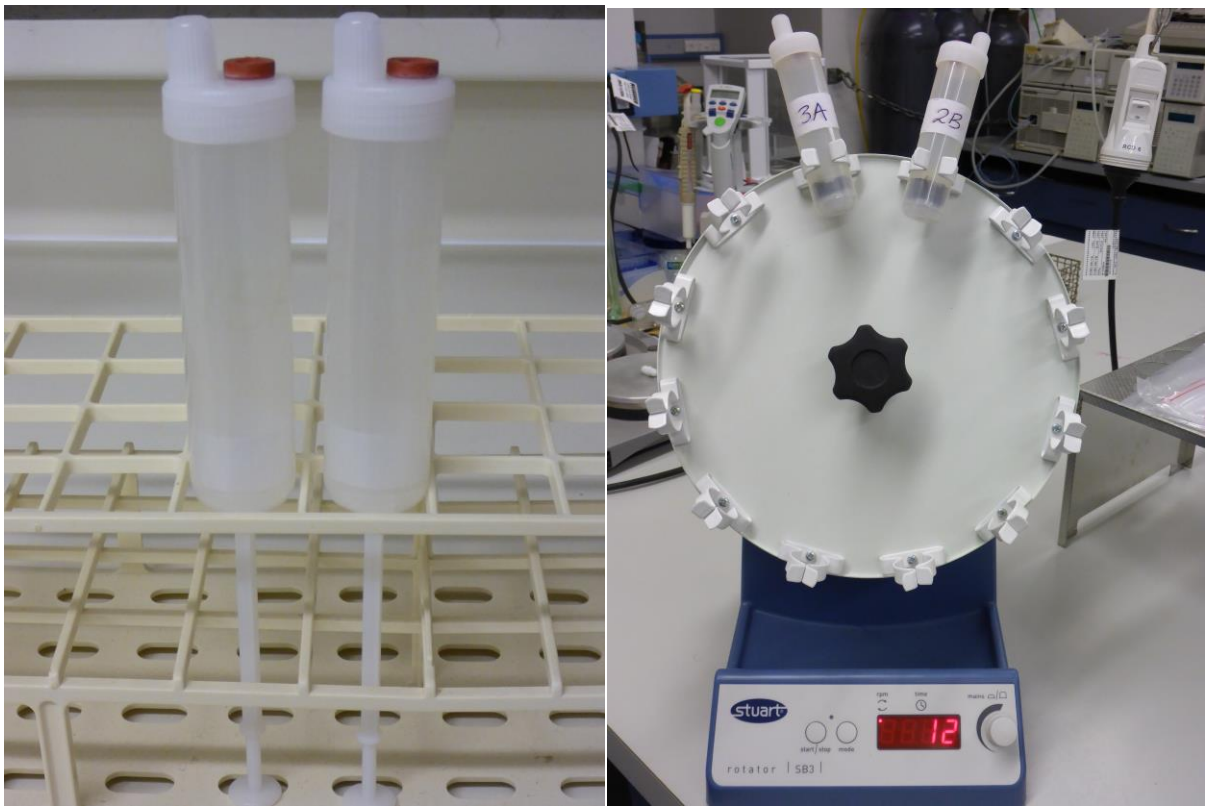
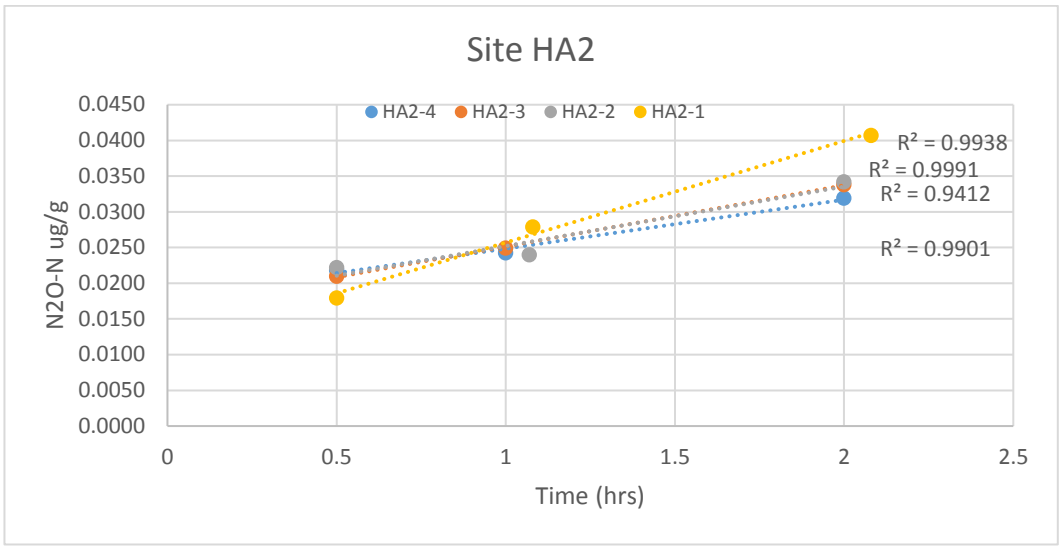


Figure 2. Monovette used for DEA assays. LHS shows the empty monovette with rubber septa used to introduce and sample gases. The soil receptacle can be seen at the bottom of the monovette with the removable plunger. The RHS shows monovettes on the shaker and the soil sample with purged NO_3-N added can be clearly seen relative to the headspace (O_2-free nitrogen with ~1% acetylene). Note the soil level would not normally be seen as the monovettes were shaken vigorously prior to being placed on the shaker.

3. Results

Examples of N_2O accumulation are shown in Figure 3 for sites HA2 and HA11 (Jones Rd).



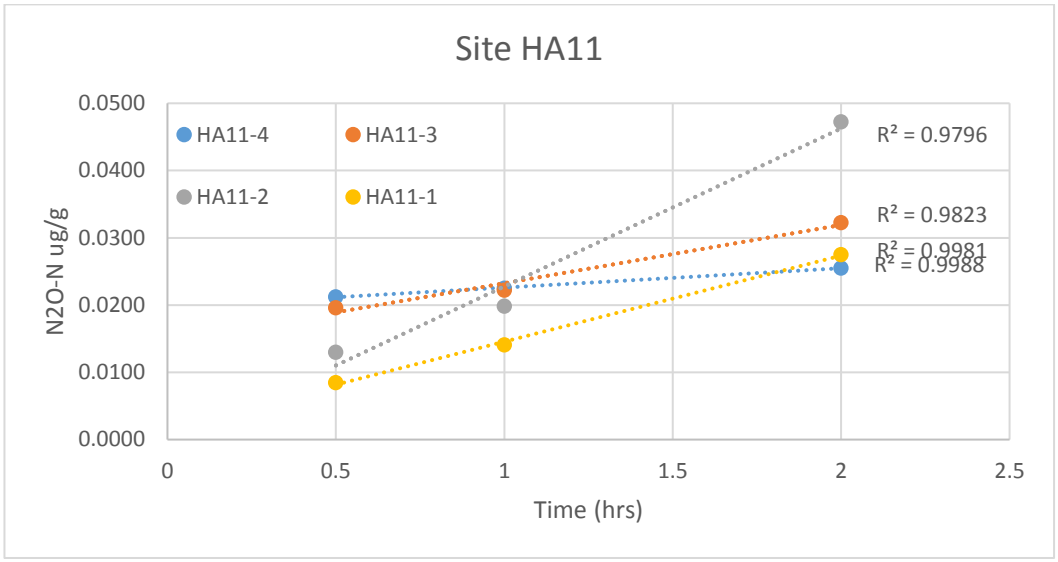


Figure 3. Examples of nitrous oxide accumulation in monovettes.

In general R^2 values were >0.98 and the DEA rates were determined from the slope of the line. The standard error of ±8.8% of the mean DEA rate was determined from a triplicate sample at the same site/depth.

The measured DEA rates for the Jones Rd and Golf Course sides of the harbour are shown in Figures 4 and 5, respectively. To enable comparison between sites, the rates are scaled relative to the highest rate measured[1] on each side (i.e. 70 ng N/g/h at Jones Rd and 35 ng N /g/h on the Golf Course side.

[1] The exception is HA5, which is discussed in text.

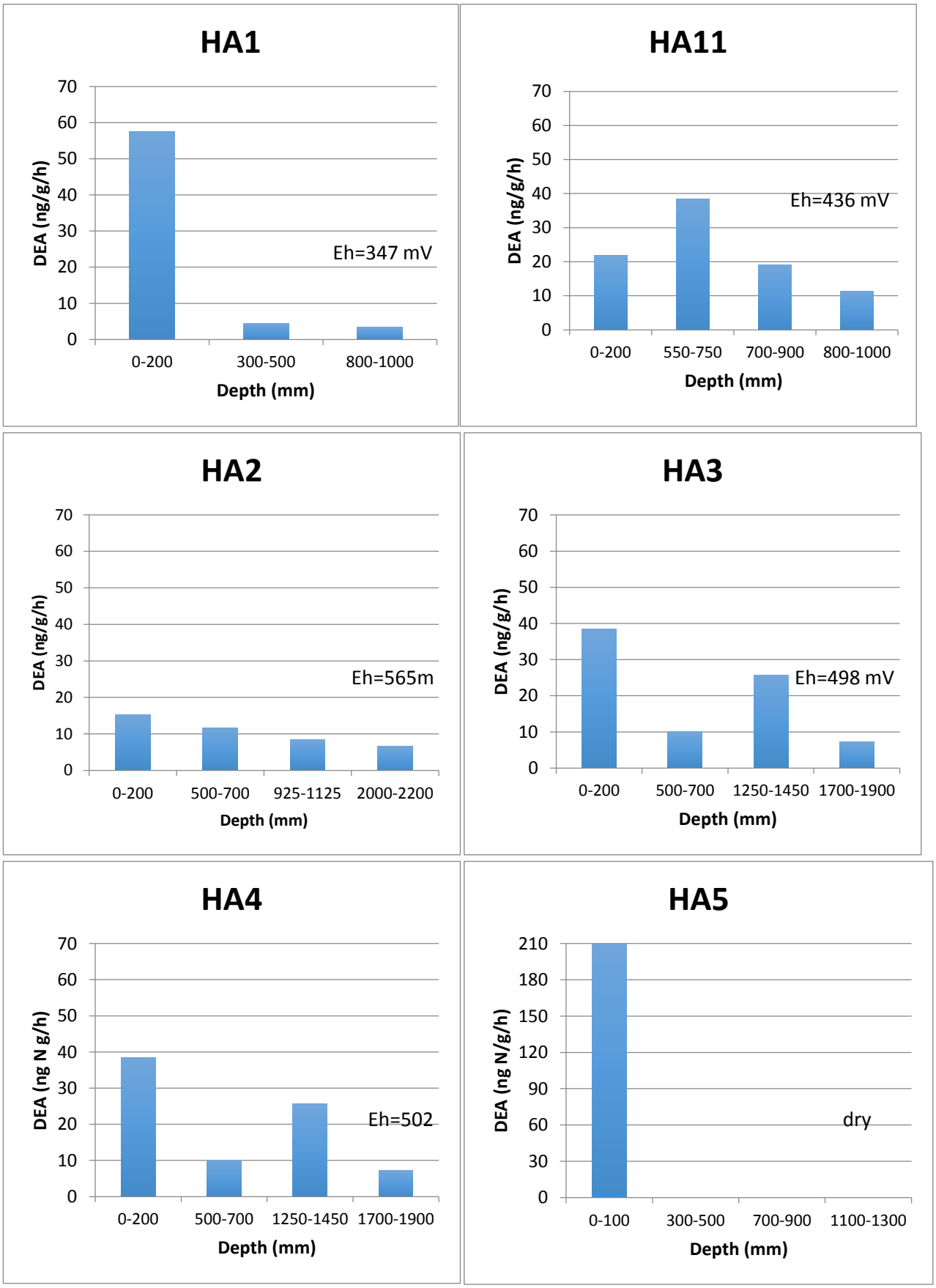


Figure 4. DEA as function of depth at Jones Road site (note change in scale for HA5).

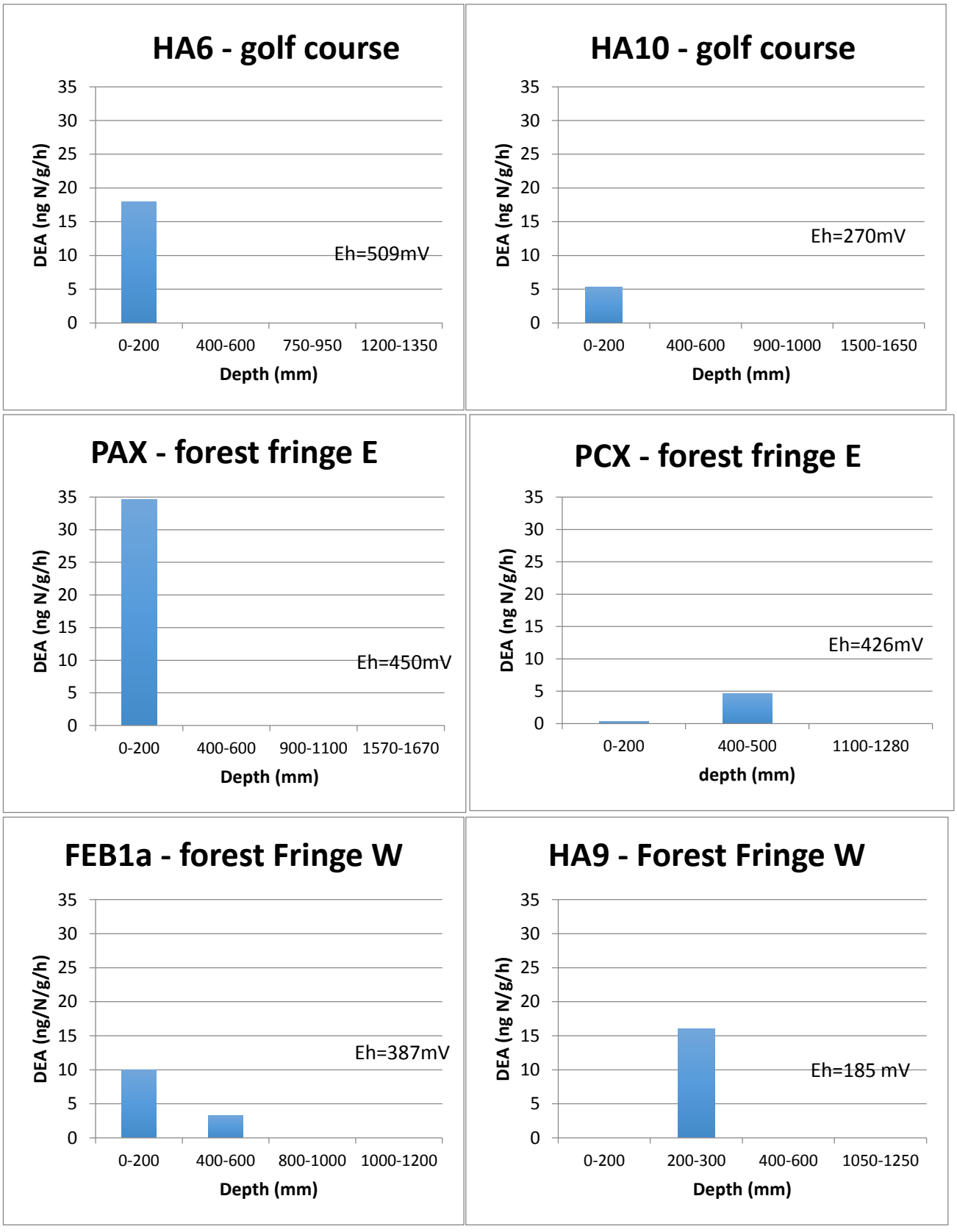


Figure 5. DEA as function of depth at eastern side (Golf Course) of Whangateau Harbour. Note - absence of bars indicates zero DEA recorded. Redox potential (Eh) is shown at water table depth.

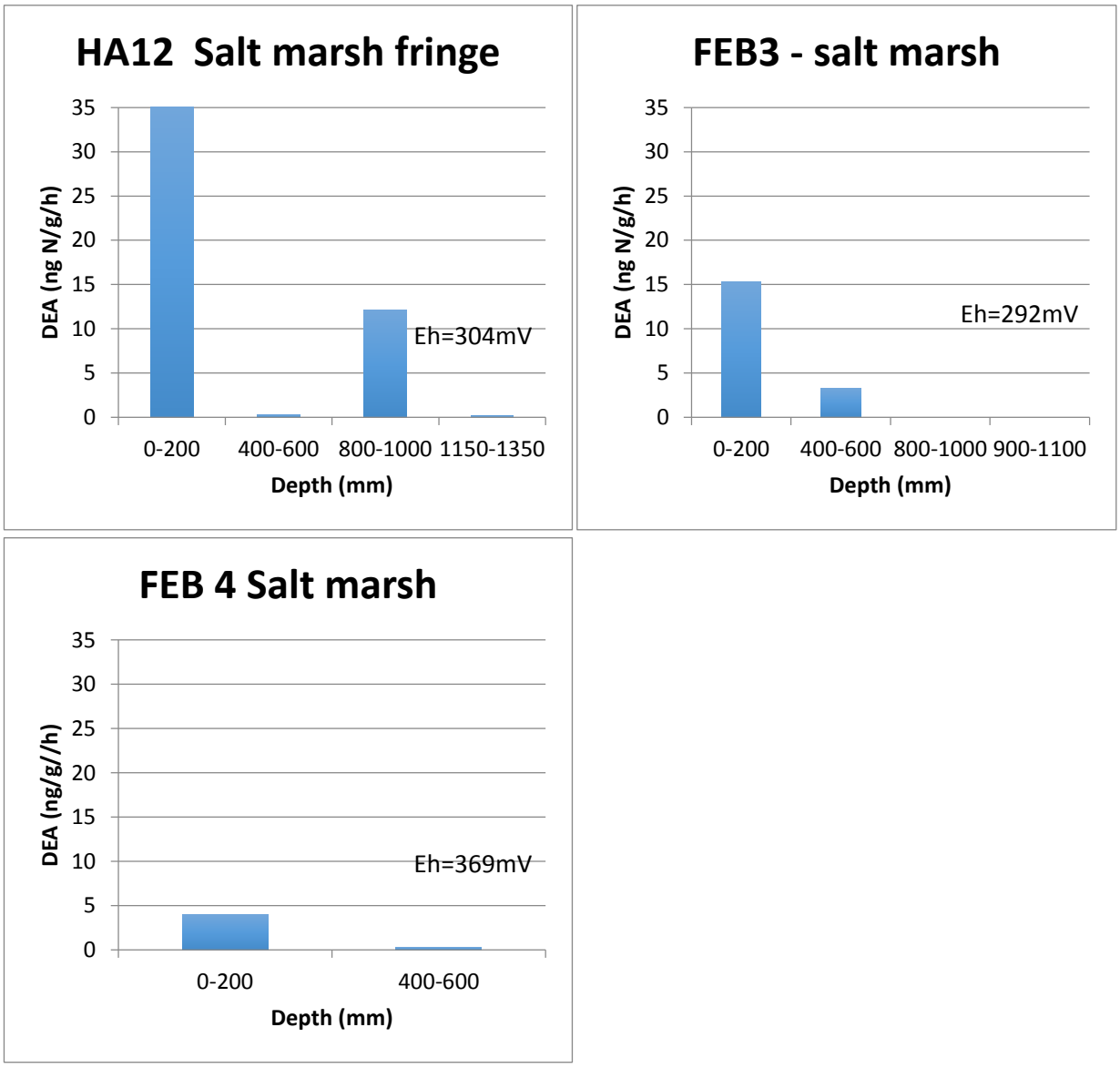


Figure 5 continued - DEA as function of depth at eastern side (Golf Course) of Whangateau Harbour. Note - absence of bars indicates zero DEA recorded. Redox potential (Eh) is shown at water table depth.

In general the highest DEAs were recorded in the topmost soil layer (0-200mm) with a steady decrease with depth. HA11 was the exception with highest rates recorded at the 550-750 mm sample. At PCX and HA9 the only activity was recorded at these intermediate depths. At the Golf Course site (HA6 and HA10) only the top 200mm had any measureable DEA. The lack of activity below 200 mm at the golf course was coincident with the profile consisting of fine-medium sand with no noticeable organic matter. In contrast there was measureable DEA at all sites and all depths on the Jones Rd side of the harbour with the exception of HA5. The highest DEA of any sample (210 ng/N/g/h) was recorded in the 0-200mm sample at site HA5, with no activity at any depth below that. As HA5 was on the airfield, the furthest away from the irrigation site and the core was dry down to at least 1.3 m, it seems very unlikely that the high activity was due to the irrigation.

Redox potential (Eh) was measured where the soil became saturated (i.e. at the water table). At all of the irrigation sites except HA10, the Eh was greater +400mV indicating oxic conditions. At site HA10 the Eh at the water table was +270mv which is around the range where denitrification might be expected to be optimum (i.e. oxygen not present, but not strongly reducing conditions where Fe or S reduction might be expected. The sites downslope of the Jones Rd irrigation sites (airfield HA4 and HA5) were still strongly oxic, as were the sites immediately downslope of the golf course irrigation (PAX and PCX). At the far side of the forest, however, the Eh at the water table was lower (+185mV-387mV), as were the salt marsh sites (+292-369 mV) which is more in the range considered optimum for denitrification (+10-300mV).

4. Discussion

The measurement of DEA is a surrogate indicator of recent denitrification, such as might occur when a high nitrate-containing leachate (from irrigated wastewater) passed through a zone where denitrifying microorganisms were present, and conditions were not limiting to denitrification (i.e. there is sufficient available organic matter to sustain the energy requirements of the denitrifiers, and oxygen is absent. Under such conditions nitrate becomes the limiting factor for denitrification.

Denitrifying microorganisms are fairly ubiquitous in nature, i.e. a large range of genera has denitrifying ability. However they are classed as facultative aerobes, which means that when oxygen is present they will preferentially use oxygen for their metabolism. It is only when oxygen is absent, and nitrate is present that they use NO_3^- as a terminal electron acceptor and denitrification proceeds.

The results of this study indicate that at many sites either: (i) the depths selected were not in continuous contact with a high nitrate source (such as might be expected from a continuous leachate field), and/or (ii) the conditions were not conducive to denitrification (lack of organic matter, presence of oxygen). It is likely that both these conditions existed at the Omaha WWTP irrigation sites, though usually not at the same site/depth combinations. Certainly at the golf course, whilst there is probably adequate nitrate at the time of irrigation, the combination of free draining sandy soils and lack of organic matter below about 200mm is not conducive to denitrification and it is therefore not surprising that no activity was detected at these depths. Adjacent to the golf course at the forest fringe (sites PAX and PCX) similar conditions exist, and it is possible that the leachate from the golf course irrigation passes underneath the deepest sites sampled (~1.5 m). On the other side of the forest (FEB1a, HA9) conditions appeared more suitable for denitrification (low oxygen, presence of organic matter) but the question remains, does the nitrate containing leachate actually intersect the sites sampled? Certainly there was measureable DEA, which indicates that denitrifiers are present, but we cannot be sure that the source of the activity is the irrigation field. Similarly, out in the salt marsh, the conditions are favourable for denitrification if nitrate-containing leachate intersects the site. Again there was measurable activity, which indicates that denitrifiers are present and will reduce nitrate providing oxygen is absent. It is likely in this environment, such conditions will occur more of the time than is case at the eastern forest fringe. We had no sites within the Kahikatea Forest itself, which is where there is considerable accumulation of organic

matter and DEA might be expected to be high provided there was a nitrate source present (Diffuse Sources, 2008).

On the Jones Road irrigation site we have more confidence that denitrification is occurring at the irrigation site. Whilst the redox measurements at the water table indicate oxic conditions (unfavourable for denitrification), it is clear there is abundant organic matter and that within the peat making up the bulk of the organic matter, diffusion/consumption processes would make oxygen limiting. In such a scenario with both oxygen and nitrate dissolved in soil water, nitrate will diffuse further (Cooke and White, 1987) and a zone of active denitrification will occur. The fact that DEA was measureable at all depths on the Jones road site (albeit at low rates for some sites) gives credence to this conclusion. Offsite, however, the picture is less convincing. At HA4 (airfield) there was measurable DEA at all depths and there were peat layers within the profile. At HA5 however very high rates were measured at the top depth, but zero activity beneath this depth. It does not seem plausible from the soil profiles that leachate from irrigation field would emerge close to the surface at HA5, and it is more likely that the high activity at the surface is a consequence on recent fertilization and or defecation (horses grazed adjacent to the site).

The results of this study are consistent with those of Barton et al. (2000) on the Rotorua land treatment system. On the coarse-textured free draining soils there they concluded that size of the soil denitrifying population in the Land Treatment System appeared to be limited by soil aeration, and that size of the denitrifying population cannot be expected to be large in free-draining, coarsely textured soils even when provided with additional nitrogen and water inputs. It was only where the leachate from those soils (and hydraulic wastewater loading was very much greater than that at Omaha) drained into organic rich wetlands that significant denitrification occurred.

The situation at Omaha is complex. On the golf course site conditions are generally not suitable for denitrification at the irrigation site itself, but are potentially suitable in the Kahikatea forest adjacent to it, and the salt marsh beyond that. At the Jones Road site, conditions are generally favourable for denitrification onsite, but generally unfavourable offsite (though questions remain in terms of frequency of peat lenses and whether leachate or groundwater intersects these lenses. Compounding this uncertainty from a scientific perspective is the variable nature of the irrigation schedules, the cycling between irrigation sites at the golf course, and climatic influences (which had a marked effect on depth to groundwater even in the month between PDP's hydraulic survey and our survey).

5. Recommendations

In the revised investigation plan (November 2014) a second phase of the nutrient processing work planned to resample the sites for DEA and do selective in situ denitrification measurements on the irrigation sites. Other work was planned around measuring redox potential, readily mineralisable carbon and dissolved organic carbon at bore sites.

In view of the results above, our view is that this plan should be revisited and predicated on a very good understanding of the hydraulic pathways from the irrigation sites to the harbour. Without such an understanding, the measurement of DEA, redox potential, mineralisable carbon etc designed to improve our understanding of nitrogen removal is without context, since we have no certainty that leachate actually passes

through the sampling zone. The sampling sites for this (Phase 1) study, were selected primarily to calibrate a groundwater model. We recommend that selection of sites away from the irrigation field itself, should be based on a successful validation of this model.

There is scope in our view for further phase 2 work at the irrigation sites themselves. The sites selected in this phase 1 study were not optimal, with the Jones Road sites being at the end of irrigation lines rather than immediately downslope. Similarly the golf course sites were in 'rough' away from the fairways that are the target areas. Although these 'rough' sites are downslope of the irrigation lines, given the sandy nature of the soils it is quite possible that leachate passes beneath the sites sampled. In order to get estimates of the maximum rates at the irrigation sites, it would be better to select sites that we can be certain have been irrigated at the end of an irrigation cycle. There may also be benefit in considering deploying suction lysimeters at these sites to obtain data on the fate of N in an irrigation cycle as it passes through the soil profile. At the Jones Rd site it may be that more or less complete N removal is effected at the irrigation site itself (we calculate that at the DEA rates recorded in this phase 1 study, < 2 ha of land would be required to effect complete N removal at the current application rates). Collecting data to test whether this is actually the case in practice would be useful.

6. References

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