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University of York

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REPORT ON CURRENT ECOLOGICAL STATUS OF HESLINGTON LAKE AND A PROPOSAL FOR AN ONGOING MONITORING PROGRAMME

ABSTRACT

Macrophytes, diatoms and chemical indicators were studied to evaluate the water quality of Heslington West Lake, to assess whether the lake met the WFD 'Good Status'.

EQR values were conducted to determine ecological status classes as required by the EU Water Framework Directive. Analsysis revealed that phytobenthos had an EQR value of 0.99 suggesting that the water quality is high. However, no macrophytes were collected so an EQR value wasn't calculated.

A lack of macrophytes is a possible reflection of differences in lake quality and local pollution, however chemical indicators measured in the lake were similar to the surrounding water bodies, suggesting that the lack of macrophytes is not necessarily effected by the water quality but instead it may be effected by sampling time, shade, light availability or calcium and magnesium concentrations.

Due to the uncertainty of the lakes current status a proposed monitoring plan has been suggested in order to establish the ecological status of Heslington West Lake, with details concentrating on monitoring times and frequency.

1. INTRODUCTION

The Water framework directive (WFD) aimed for all surface waters to reach 'good status' by 2015, to help maintain and improve the European unions aquatic environments (Boeuf & Fritsch, 2016). It's important to manage, maintain and support the diverse biotas that lakes have (Vadeboncoeur *et al.,* 2011) as biodiversity and ecological status' of lakes are threatened by climate change, degradation of habitats, invasions of alien species, eutrophication, pollution (Vadeboncoeur *et al.,* 2011) and waterfowl (Birch & McCaskie, 1999).

To ensure that 'Good status' is being met it's important to conduct water quality studies. Suitable methods of water quality assessment include assessing macrophytes and phytobenthos as they're ecological indicators that act as signals for stresses effecting lake ecosystems (Rapport, 1995). This is largely due to water conditions being outside the range of tolerance of which some organisms can flourish, suggesting that organism distribution is a direct result of the variation in the quality of the water (Hellawell, 2012). As biological indicators are exposed to variations in the quality of lakes and the environment they are more sought after sampling methods due to their ability to reflect the peaks, troughs or sustained levels of quality (Hellawell, 2012).

Similar methods will be used to assess the status of our study site as it faces similar pressures and threats to other lakes. Heslington West Lake was constructed in the 1960's and is Europe's largest plastic bottomed lake (University of York, 2018). The lake was developed to act as a balancing reservoir for the surface water drainage, as increased building developments would have overloaded the existing drainage system with runoff (University of York, 2013). The pressures from leaf litter, ground water run off, water fowl and geese who create a large amount of excreta, were neglected during the development of the lake so the lake now faces issues of nutrient enrichment, unbalanced ecosystems and aeration (University of York, 2013). In 2013 Heslington West Lake the water quality was degraded and didn't fulfil it's potential to support a variety of ecosystems (University of York, 2013).

Therefore, the purpose of this study is to determine whether the University lake achieves the 'Good status' that the water framework directive sets out to achieve. Based on the report findings, a monitoring programme is going to be proposed that could be applied for an ongoing assessment of the ecological status of the lake.

2. METHODOLOGY

2.1 STUDY AREA

Heslington West Lake in the grounds of the University of York and has a mean depth of 1.2m (additional 3000mm of silt accumulation on the lake bed), an approximate surface area of 5.34 ha (University of York, 2013), an expected alkalinity of the lake is 105 mg/l as this is the recorded value for the Heslington area (Yorkshire Water, 2017) and is at an altitude of 14.02m.

A general lake survey was conducted were descriptive notes were taken (**Appendix A**), which allowed for two suitable sample sites that would represent the whole lake to be located.

2.2 SAMPLING METHODS

Several different methods were used to assess Heslington West Lake's water quality. For specific details of methods undertaken for this report see **Appendix B.**

2.2.1 SURVEY GROUPS

Shoreline sectors with a length of 100m were measured at the two sites. At every 10m section of each location the following steps were conducted:

- Recorded the presence of different emergent, floating and submerged macrophyte.
- Created a sketch map of each 10m section to record substrata, percentage cover, percentage of shoreline shaded and presence of macrophytes.
- Every two metres along the shoreline a transect was made to record depth every 5cm from the shoreline.
- Collected Pebbles/ boulders/ cobbles to analyse in the lab.

2.2.2 WADING GROUPS

Wading groups conducted transects at the following intervals along the 100m shoreline: 10m, 30m, 50m, 70m, 90m, and took the following steps at each interval:

- Made a transect to record depth every 5cm from the shoreline
- Recorded levels of dissolved oxygen levels and water temperature
- Collected pebbles/boulders for laboratory analysis
- Collected water samples from just below the waters surface

2.3 LABORATORY ANALYSIS

Laboratory analysis was undertaken for all samples of water and substrates collected in the field.

2.3.1 SOLID SUBSTRATES

Pebbles, cobbles and boulders collected were scrubbed vigorously with the solution being placed into a centrifuge tube. Samples were then centrifuged at 2500rpm for 10 minutes. The top 40ml of supernatant was poured off, leaving approximately 10mls of liquid. Samples were poured together and shaken to give a thick suspension.

2.3.2 MACROPHYTES

No macrophytes were collected from the field.

For details on what method would have taken place if macrophytes had been collected please see **Appendix C.**

2.3. WATER SAMPLE ANALYSIS

Two sets of 5ml of lake water were filtered into two 15ml centrifuge tubes using a 0.2um Whatman filter. One sample was analysed for metals, whilst the other sample was used to analyse anions. The remaining unfiltered samples were also analysed for pH, conductivity, alkalinity, nitrate and phosphate levels. Water sample analysis was conducted using the equipment in **Table 1**.

Table 1 Equipment that was used to test the different Environmental variables in the collected water samples

Environmental variable	Units	Equipment
pH	pH units	Accument AP72 pH Data Meter
Conductivity	µS/cm	HI 9033 Multi-Range Conductivity meter
Alkalinity	Mg/l	HI 3811 Alkalinity Test Kit
Nitrate	Mg/l	HACH Aquachek Water Quality Test Strips for Nitrate
Phosphate	Mg/l	Quantofix Phosphate Test Kit
Anions	Mg/l	Dionex ICS2000 Ion Chromotograph
Metals	Mg/l	Inductively-Coupled Plasma Optical Emission Spectroscopy: Thermo Scientific iCap 7000 series ICP spectrometer

2.4 PHYTOBENTHOS IDENTIFICATION

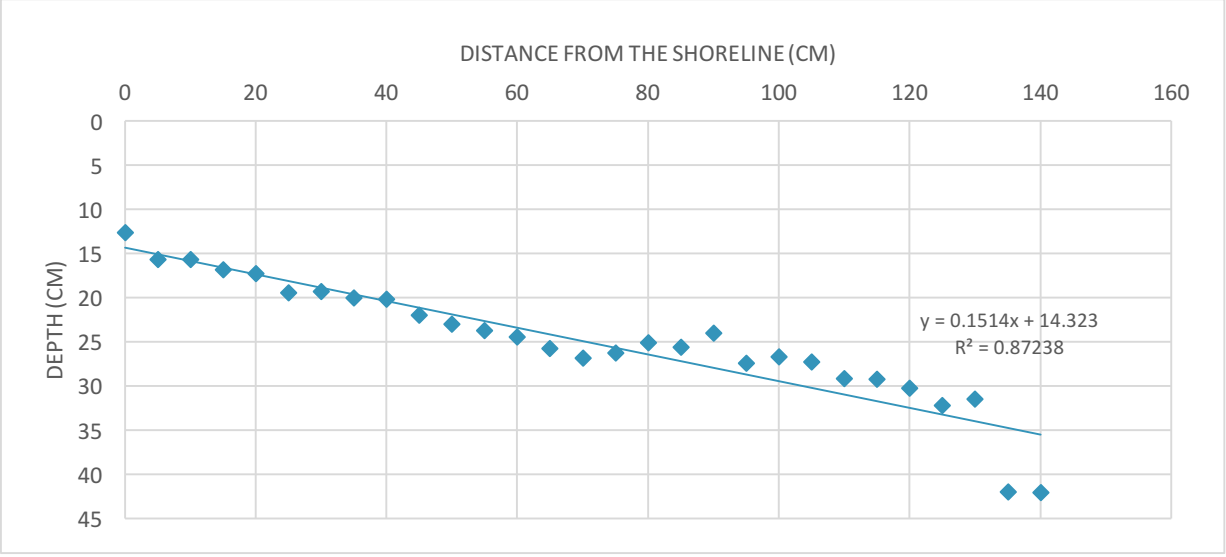
Diatom samples were cleaned using Hydrogen peroxide and mounted on permanent slides that had been prepared using Naphrax as the diatom mountant. Slides were examined using a Nikon Eclipse e200 microscope under x1000 magnification with a small drop of immersion oil. Diatoms identified under the microscope were identified to genus level with the number of each taxa observed in the field of view being counted. EQR values were then calculated using the UKTAG guidelines. For the calculations used see **Appendix D.**

3. RESULTS:

3.1 PHYSICAL DESCRIPTION OF THE LAKE

The lake profile was derived from the depth measurements collected by the wading teams (**Appendix E**) rather than the survey teams due to human error in those data sets. The lake slope calculated a gradient of m=0.1514, suggesting that slope angle is minimal and that the lake does not deepen rapidly but is gradual as displayed in **Figure 1**.

Figure 1 A graph showing the calculated slope gradient of Heslington West Lake



3.2 WATER QUALITY ANALYSIS

Analysis of the raw data (**Appendix F**) revealed the abundance of the major anions in the order Chloride> Sulphate> Nitrate> Nitrite> Phosphate as shown in **Figures 2** and **3.**

Figure 2 A graph displaying the average levels of Anions Chloride and Sulphate in Heslington West Lake

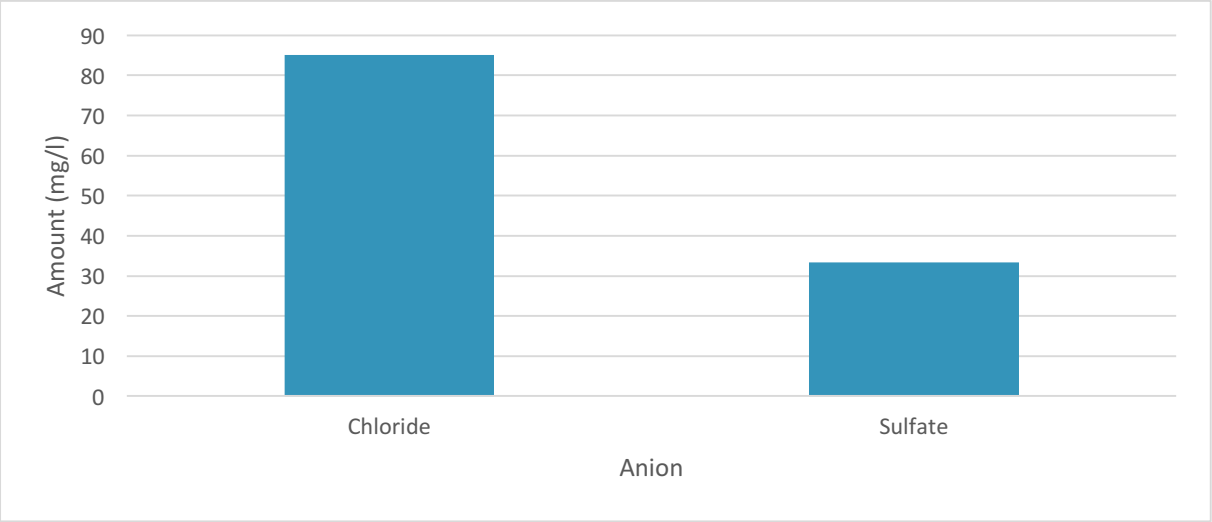
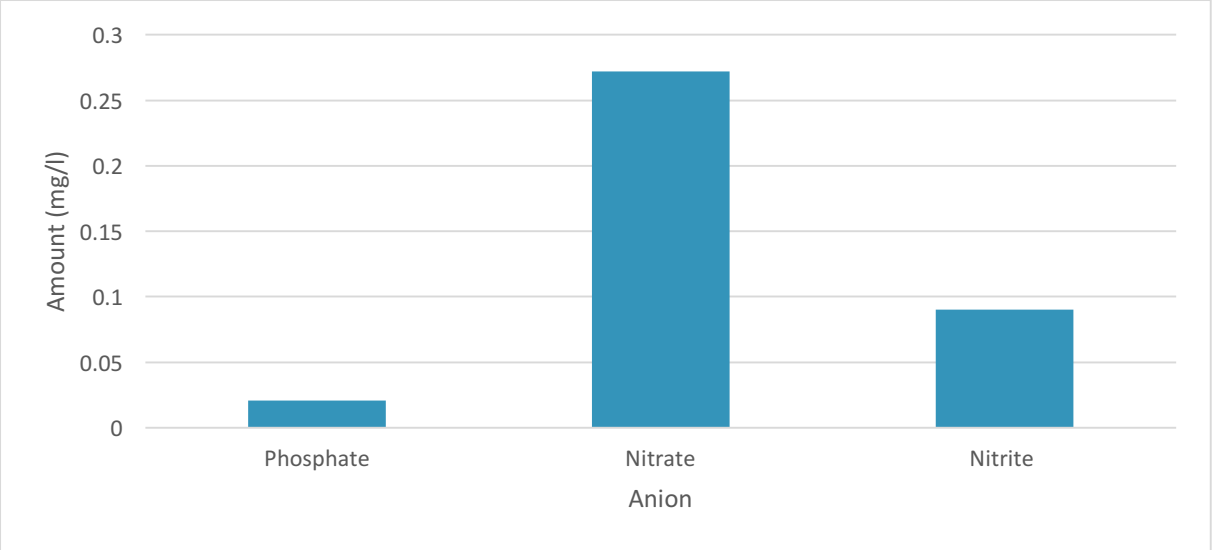


Figure 3 A graph displaying the average levels of Anions Phosphate, Nitrate and Nitrite in Heslington West Lake



From the raw data in **Appendix G**, further analysis unfiltered water samples were conducted with results displayed in **Table 2**. Conductivity had readings ranging from 500 µS/cm to 601 µS/cm, whilst alkalinity had readings ranging from 98 to 156 mg/l, suggesting that there may be factors effecting indicator readings.

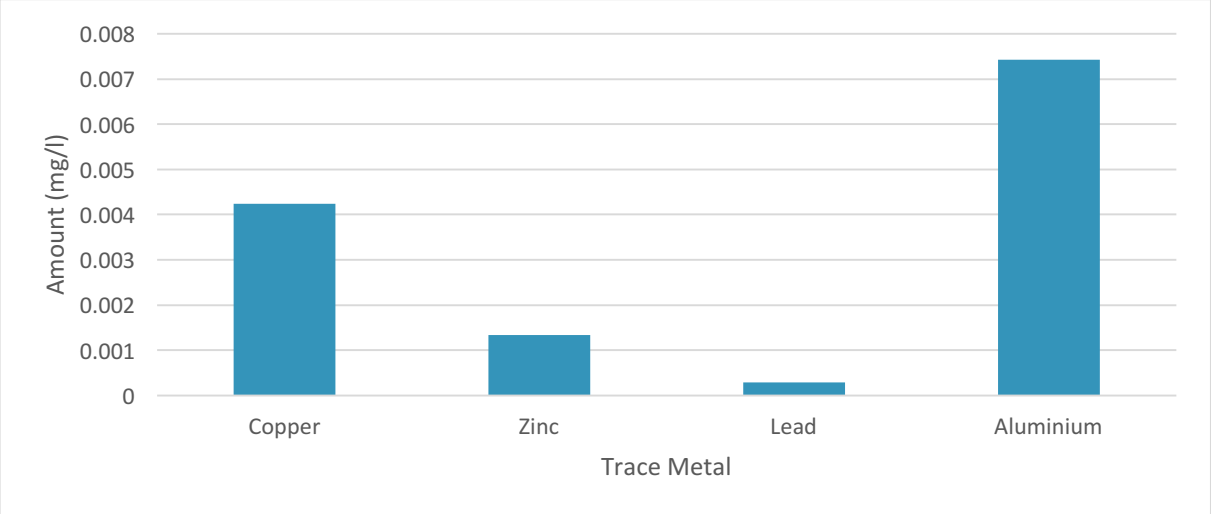
Dissolved oxygen levels varied between the two sample sites, as site one had an average reading of 3.7 mg/l, whilst site two had an average of 11.34 mg/l, suggesting there were variables at each site that influenced dissolved oxygen levels.

Table 2 Statistical results for Environmental variables tested to assess water quality

Variable	Mean	Standard deviation
Phosphate(mg/l)	1.5	±1.643167673
Nitrate concentration(mg/l)	0.8	±1.691481928
Conductivity(µS/cm)	564.8	±35.38298398
Alkalinity(mg/l)	130	±20.60204952
pH	7.694	±7.694
Dissolved oxygen (mg/l)	7.5	±4.05
Temperature (0^c)	13.66	±1.49

Analysis of the raw data (**Appendix H)** revealed that Aluminium had the highest average reading of 0.007mg/l, whilst Lead had the lowest average levels of 0.0002 mg/l, suggesting that levels of trace metals are relatively low. Results of all metals tested for are displayed in **Figure 4**.

Figure 4 A graph showing the average levels of Trace metals found in the water at Heslington West Lake



3.5 MACROPHYTES & PHYTOBENTHOS

A substantial number of diatoms were identified, however, due to the misidentification of diatoms some data sets was excluded as seen in red in **Appendix I.** The diatoms identified ranged in sensitivity scores, from 0 to 5, however, a large proportion of the data was awarded a sensitivity score of 0 as they weren't found in the appendix of the UKTAG guidelines. Diatoms not found in the appendix were kept in the data set and given a score of 0 as they were clearly identified in the samples. However, this could potentially have had an impact on the calculated EQR value.

The EQR calculated revealed that water quality in Heslington West Lake was high, as the EQR value calculated was 0.99. Raw data and calculations of the EQR value can be seen in **Appendix J**.

No macrophytes were collected in the field due to a lack of macrophytes in the field that appeared on the Appendix of the UKTAG guidelines, therefore no EQR calculations could be made for water quality. However, site Surveys revealed several different macrophytes species at site one (**Table 3**). The macrophyte species all varied in location along the 100m transect, depth from the shoreline and percentage of cover (**Appendix K**). The overall percentage cover of macrophytes at site one was calculated as 18.9% of the 100m transect.

Table 3 Macrophyte species present at site one

Species name	Scientific name
Common reed	*Phragmices australis*
Reed Sweet Grass	*Glyceria maxima*
Bulrush	*Thypa latifolia*
Large Rush	*Juncus effusus*

4. DISCUSSION:

It's important that lakes achieve 'Good Satus' as it allows for all aspects of a lake to be sustainable and effective (Teodosiu *et al*., 2003). Water quality analysis using biological and chemical parameters provided evidence that Heslington West Lake reaches 'Good status' in terms of phytobenthos, however due to a lack of data, an

EQR value for macrophytes was not conducted. Although macrophytes were present in the lake, they were not in the UKTAG guidelines, which suggests that environmental variables may be impacting upon macrophyte growth.

4.1 ENVIRONMENTAL VARIABLES

Dissolved oxygen plays a vital part as it's essential to a lakes overall water quality (Sharifinia *et al*., 2013). Dissolved oxygen varied between site one and two, with levels being lower at site one. However, this could be linked to the significant amount of detritus present, as the decomposition process requires a significant demand of oxygen (Sharifinia *et al*., 2013). Considering the difference in readings due to different site factors, the overall average of 7.5 mg/l was similar to a study conducted by Sharifinia *et al.,* (2013), which found an average reading of dissolved oxygen of 7.29 mg/l. The recommended guidelines of dissolved oxygen levels are 5 mg/l, as anything under this can cause stress to aquatic life (Lenntech, 2018).

pH is detrimental to bacterial community composition, so it's one of the most important factors of water compared to temperature, nutrient concentrations and water retention time (Lindstrom *et al.,* 2005, Newton *et al.,* 2011, Ren *et al.,* 2015). Therefore, it's essential that pH levels of Heslington West Lake are kept between the recommended guidelines of 6.5-9.5. pH levels recorded at Heslington West Lake are within the recommended guidelines with a pH that's similar to water bodies in the surrounding areas (**Table 4**). It's important to understand that pH fluctuates rapidly (Baath & Kritzberg, 2015) as it's effected by the surrounding rock, precipitation, waste water and sewage discharges (Fondriest Environmental, 2013). It's been found that during intense rain storms and snow melt, pH can decrease by several units (Wigington *et al.,* 1992), or in events of warm, sunny weather when photosynthesis is high, pH increases (Baath & Kritzberg, 2015).

For water quality to be deemed 'Good status' it's essential that trace metals are present in lake waters as they're necessary for metabolic and enzyme reactions that occur in organisms (Lehninger, 1982). The levels of trace metals meet the expected levels for the Heslington area along with conductivity levels which is also similar to the expected readings (**Table 4**), suggesting that conductivity and trace metal levels will not directly effecting the lakes current status.

It's important that conductivity levels and trace metals are similar to those of the surrounding areas, as many organisms do not have the tolerance levels to cope with large fluctuations in conductivity and trace metal levels (Fondriest Environmental, 2014).

The average Alkalinity measurement of Heslington West Lake was above the value normally recorded in the Heslington area (**Table 4**). Although higher than the normal recorded values, a high alkalinity in water is preferable for many aquatic organisms, as this allows for a stable pH value (Addy *et al.,* 2004), so it is not likely to have major impacts on the ecological status. High alkalinity allows for water to neutralize any potential acidic pollution, as well as acting as a lake buffer so that fluctuations in CO_2 only present minor changes to pH, however alkalinity is potentially being influenced by road salt in the winter months (Addy *et al.,* 2004).

Table 4 Typical values of chemical and environmental parameter found in Heslington water (Yorkshire Water, 2017) and the actual data recorded during assessment of Heslington West Lake

Substance	Typical Value for Heslington	Value recorded in Heslington West Lake
pH	7.3	7.8
Alkalinity	105 mg/l	130 mg/l
Conductivity	571 µS/cm	564.8 µS/cm
Lead	0.00408 mg/l	0.000286655 mg/l
Aluminum	0.0086 mg/l	0.007428211 mg/l
Copper	0.0068 mg/l	0.004243591 mg/l

4.2 CHEMICAL COMPOSITION

The main chemical composition of freshwater lakes is sulphate, chloride and nitrate (Tchobanoglous & Schroeder, 1985). Chloride in freshwater lakes is usually between 0-100 mg/l, but can be effected by low flow periods during the summer, when evaporation exceeds precipitation, or more recently by anthropogenic activities, including sewage concentrations and road salt (Hunt *et al*., 2012). It's most likely that the high levels of chloride in Heslington West Lake is a result of the warm summer weather that the UK experienced, as road salt is applied in the winter months.

Chemicals in lakes such as sulphate and nitrate varies depending on season (Lane, 2018), suggesting that sulphate and nitrate levels might increase during the winter months due to increased rain, run off and ground saturation (Kerekes *et al.,* 1986).
Although chemical composition in lakes is clearly effected by different factors, it's not clear whether chemical composition in Heslington West Lake is specifically effected by the factors mentioned. Therefore, further monitoring is required to see if chemical composition is effected by seasonal variations.

4.3 INDICATORS OF WATER QUALITY

As no macrophyte EQR value was calculated the status of the lake could not be determined. However, it doesn't mean that water quality was poor as some macrophytes species were found. The 4 macrophytes species found are common across the UK and often appear in wetland areas, so do not appear on the UKTAG guidelines appendix. It can't be assumed that water quality is poor due to the lack of macrophytes, as sampling was conducted at the end of September when macrophytes begin to die as seen by the detritus at site one.
Studies in shallow freshwater lakes found that environmental variables including woodland cover, pH, calcium and magnesium concentrations and alkalinity influence macrophyte distribution (Heegard *et al.,* 2001; Kissoon *et al.,* 2013). Alkalinity and pH were found to be lower at site one where macrophytes were found, whilst several areas of each site were either completely shaded or had partial shade which reduces light, with only certain species being able to tolerate low light (Stuckey, 1975). It suggests that these environmental variables may impact upon the macrophyte distribution in Heslington West Lake rather than the water quality.
Water quality in urban lakes can change rapidly as a result of polluted run off, construction work and road salt, which often override factors like water chemistry (Choe *et al.,* 2002). Diatoms in water can respond quickly to pollution events especially if events effecting conductivity, pH and nutrients (Guzkowska & Gasse, 1990). This assessment suggests that Heslington West Lake has had minimal pollution events occur due to the strong presence of different diatoms with a range of sensitivity. Although the results implicate high water quality due to the different diatoms, this could mainly be a result of human error due to the groups lack of experience in assessing diatoms, as several sets of data did have to be removed due to incorrect identification.

However, the high EQR value may also be a result of diatoms only decreasing when pollution is severe, (Jüttner *et al.,* 2003).

5. CONCLUSION

Diatom and macrophytes pose as valuable indicators of water quality in urban lakes (Jüttner *et al.,* 2010) such as Heslington West Lake, due to their ability to reflect changes to water quality from events such as localized pollution (Passy, 2001).

This assessment revealed that Heslington West Lake had a high EQR value, with diatoms indicating that the lake had a high ecological status. This suggests that Heslington West Lake achieves the 'Good Status' put forward by the WFD. However, there was no macrophyte EQR calculated, which suggests that the water quality is not as high as the diatoms suggest. There are a variety of factors that may influence the lack of macrophytes and the high abundance of diatoms throughout the lake, however due to the assessment being conducted over one day it doesn't show the potential changes to the water quality over a period of time which may effect biological indicators. It's therefore essential that continued monitoring of Heslington West Lake occurs to see if changes to environmental variables, seasonal variations and water chemistry effects diatoms and macrophytes in the future which could eventually effect the water quality status of the lake.

6. MONITORING PROPOSAL:

The assessment methods used revealed that there wasn't a clear understanding on Heslington Lake current ecological status. Methods used weren't robust enough as assessment was carried out in one day and there was a lack of understanding amongst the group, which lead to human error in several data sets. Potentially these issues may be the main cause of such a high EQR value being produced by the diatom calculations, but studies have also found that diatom richness is a poor indicator to use for water quality as it only declines significantly when there is a severe pollution event (Briggs & Smith, 2002), suggesting that more focus should be aimed towards macrophytes and chemical indicators.

Therefore, the following improvements can be made to the current monitoring methods used in this assessment (**Table 5**), so that water quality can be assessed through a chemical and biological data.

Table 5 Potential monitoring programme that could be used to assess Heslington West Lake's ecological status

	Details	Explanation
Sampling Design	Frequency	3 times between May and October to cover the seasons Spring, Summer, Autumn (Elias *et al.,* 2008), so samples can reflect seasonal changes. This would particularly help the with the seasonal succession of diatoms as they can be effected by high surface run off (Jüttner *et al*., 2010). As the sampling frequency is 3 times a year monitoring should occur every year for years, as many variables many not change a considerable amount in a short period of time (IAFG, 2015).
	Location	As a baseline survey was conducted of the site before this assessment commenced, it is appropriate that the two sites used in the assessment of Heslington West Lake were adequate for future monitoring purposes. These areas will be the routine location for measuring all water quality indicators.
	Depths	Sampling at multiple depths can work best for calculating the water quality indicators of the whole lake (Elias *et al.,* 2008), however as Heslington West Lake has a mean depth of only 1.2 metres, so this cannot be applied. For Heslington West Lake it may be beneficial to continue sampling at the same depths as we did for this assessment.
	Timing	Sampling of Heslington West Lake, should try and be made at the same time of day for every visit, as this will allow for variation to be minimized.
	Replication	Sampling should be more visits over a season rather than increasing replication on a sampling date (Elias *et al.,* 2008). However as sample collection over two 100m transects should remain, every 1m should be sampled instead of every 2m.
Sampling methods	Baseline survey	A baseline survey of the whole lake should be made, to assess any changes to the surrounding areas which might influence results. This should record weather, cloud cover, temperature and time of day that survey was conducted.
	Details of sample collection	Survey two sites with two 100m transects. Lake depth from the shoreline should be measured every 1m along the transect until the lake reaches 50cm depth. Substrata should be collected to assess diatoms. Water sample collection should continue as normal with one sample for every 20m sector.
	Macrophyte surveys	Spaced at 20m along the 100m sector, five transects should be taken with sampling occurring at 25cm, 50cm and 75cm depths. At each sampling point the following should be recorded: species present, estimate of total vegetation abundance, estimate of algal abundance (IAFG, 2015).

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APPENDIX A: SITE NOTES

DATE: 28/09/2018
TIME: 9:30AM-12:30PM
WEATHER: 14^{o}c
CLOUD COVER: No cloud cover

Site Details:

- Site is surrounded by footpaths
- Stormwater outlets
- Algae present in some of the ditches leading water to and from the lake
- Bridges
- Vehicles
- Wildfowl present
- Pedestrians
- Litter

Site one:

- Significant shading from trees for the majority of the 100m
- Bad smell
- Dying plant life
- No Wildfowl present
- Reeds and other macrophytes present
- Large accumulation of sediment

Site two:

- Little sediment accumulation
- No macrophytes present
- Less shaded than site one
- Large volumes of pedestrians pass this point of the lake
- No smell
- Water is clearer

APPENDIX B- METHOD DETAILS

Method – Field Work:

Survey teams:

1. Mark out a shoreline sector length of 100m and divide into 10 m sections. Assign two adjacent 10 m sections to each survey team.
2. Starting at the far left of their two sections (looking towards the lake) each survey team should slowly walk their first 10m section and record the presence of all emergent, floating and submerged macrophytes. Each macrophyte should be identified to the species level. If it cannot be identified in situ then remove a small sample and place in a plastic tray or bag for later identification.
3. Create a sketch map of each 10m section recording the type of substrata and an estimate of percentage cover, the percentage of the shoreline shaded (if the sun is out!) and the presence of the major stands of emergent, floating-leafed and submerged macrophytes which are then mapped on a plan of the shore outline.
4. Keep a record of both the sampling depth and distance from the shore for each stand of macrophyte.
5. Collect two cobbles or boulders or pebbles from your respective area. Gently agitate in the lake for a few seconds to remove any surface contamination and carefully place in a sampling tray with some lake water for later removal of the diatoms. Stems for emergent macrophytes can also be collected by cutting each stem above the water line and placing a plastic bottle over the remainder of the stem. Then cut slightly above the point where it emerges from the sediment and invert the bottle.
6. Repeat steps 1 to 5 for the second 10m section.

Wading team:

1. At 10, 30, 50, 70 and 90 m intervals (starting from the far left as you look at the lake) of the shoreline sector length the wading team should conduct transects. Carefully wade out to a maximum depth of 50 cm. Every 10 cm record the distance from the shore, the type of substrata and an estimate of percentage cover, and the presence of the major stands of emergent, floating-leafed and submerged macrophytes which are then mapped on a plan of the shore outline.
2. Measure the temperature and dissolved oxygen concentration, and collect a water sample from just below the surface of the water at the 50 cm depth – ensure that the bottle is completely full (free of air) before capping. Label the water sample with the survey sector and transect number.
3. Collect two cobbles or boulders or pebbles. Gently agitate in the lake for a few seconds to remove any surface contamination and carefully place in a sampling tray with some lake water for later removal of the diatoms.

Method - Laboratory procedure:

Solid substrates:

1. Gently place the sample in a sampling tray with 50 ml of lake water. Vigorously scrub the upper surface of the substratum with a small brush and then carefully decant the solution into a labelled 50ml centrifuge tube.
2. Repeat for the second sample from your respective zone for sector 1.
3. Centrifuge the sample at 2500 rpm for 10 minutes.
4. Carefully pour off the top 40 ml of the supernatant leaving approx 5-10 mls of liquid with the pellet in each centrifuge tube. Shake the samples well and pour both into a labelled 250 ml flask to give a max volume of 20ml of thick suspension.
5. Label the flask clearly with your zone and sector number and substrate type.
6. Repeat for the samples from sector 2.

Lake Water samples:

1. Using the 0.2um Whatman filters, filter approximately 5 ml of the lake water into a clearly labelled (sector and zone) 15ml centrifuge tube. Filter a further 5ml of sample into a second 15ml centrifuge tube. The first of these samples will be analysed for metals and the second will be used for analysing Nitrate/Nitrite/Phosphate/Sulfate/Chloride.
2. Using the remainder of the unfiltered water sample determine pH and conductively using the provided meters. You will also need to measure alkalinity, nitrate and phosphate levels using the provided kits.

APPENDIX C- MACROPHYTE METHOD

Macrophytes Method:

1. Gently place the sample in a sampling tray with 50 ml of lake water. Scrub with a small brush and then carefully decant the solution into a labelled 50ml centrifuge tube.
2. Repeat for each sample for sector 1.
3. Centrifuge the sample at 2500 rpm for 10 minutes.
4. Carefully pour off the top 40 ml of the supernatant leaving approx 5-10 mls of liquid with the pellet in each centrifuge tube. Shake the samples well and pool samples from 2 sector 1 macrophytes into a labelled 250 ml flask to give a max volume of 20ml of thick suspension.
5. Label the flask clearly with your zone and sector number and substrate type.
6. Repeat for the samples from sector 2.

APPENDIX D- EQR GUIDELINE CALCULATIONS FOR DIATOMS

The LTDI2 for each sample is calculated using equations 1 and 2:

Observed value of lake trophic diatom index = (W x 25) - 25

Equation 1

where:

"W" is given by the equation:

$$W = \frac{\sum_{j=1}^{n} a_j \times s_j}{\sum_{j=1}^{n} a_j}$$

Equation 2

where:

"a_j" is the number of valves of taxon j, and

"s_j" is the nutrient sensitivity score in Appendix A corresponding to the taxon represented by j.

APPENDIX E: LAKE PROFILE RAW DATA

Site one	Shoreline distance [cm]																												
(m)	0	5	10	15	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90	95	100	105	110	115	120	125	130	135	140
10	16	23	22	21	23	20	19	19	22	22	22	23	25	26	25	34	35	36	36	42	38	39	34	35	39	36			
30	2	2	2	2	2	1	2	2	3	6	6	5	4	3	3	4	5	10	6	8	6	7							
50	7	6	7	8	9	11	10	12	9	16	12	15	13	18	20	18	22	23	18	20	19	18	12	17	20				
70	6	8	9	8	7	7	10	10	10	9	10	13	11	14	15	15	17	19	20	27	23	26	33	22	17	21	25		
90	2.9	5	5.5	6	6.5	11.1	11.1	13.4	10.5	15.2	17.6	20	22	24.7	23.9	23.9	25.9	28.1	27.4	26	26.5	28.4	31.8	33.6	37	38.4	37.1	42	42.1

Site Two	Shoreline distance [cm]																												
(m)	0	5	10	15	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90	95	100	105	110	115	120	125	130		
10	27	27	25	26	26	27	27	27	25	25	29	30	30	29	31	30	31	30	32	32	34	32	32	32	33	35	35		
30	17	18	20	24	24	25	24	23	24	24	24	25	26	25	25	26	30	29	28	30	29	30	30	32	33	31	32		
50	3	10	10	16	17	31	30	30	29	28	30	28	27	28	28	28	27	25	20	29	30	31	30	31	31	32	29		
70	27	27.9	28.8	29	30.9	32	30.2	31.5	36.3	38	41	41.4	45.2	47	52.2														
90	19	30.5	27.9	29	28	30	29.8	33	33.3	36.7	38.7	36.9	42	43.2	45.8	50.3													

	Shoreline (cm)																												
	0	5	10	15	20	25	30	35	40	45	50	55	60	65	70	75	80	85	90	95	100	105	110	115	120	125	130	135	140
Site 1 Mean	6.78	8.8	9.1	9	9.5	10.02	10.42	11.28	10.9	13.64	13.52	15.2	15	17.14	17.38	18.98	20.98	23.22	21.48	24.6	22.5	23.68	27.7	26.9	28.25	31.8	31.05	42	42.1
Site 2 Mean	18.6	22.68	22.34	24.8	25.18	29	28.2	28.9	29.52	30.34	32.54	32.26	34.04	34.44	36.4	33.575	29.33	28	26.66	30.33	31	31	30.66	31.67	32.22	32.66	32		
Overall mean	12.69	15.74	15.72	16.9	17.34	19.51	19.31	20.09	20.21	21.99	23.03	23.73	24.52	25.79	26.89	26.2775	21.15	25.61	24.07	27.46	26.75	27.34	29.18	29.28	30.29	32.23	31.525	42	42.1

APPENDIX F: ANION RAW DATA

	Amount (mg/L)			
	Chloride	**Nitrate**	**Sulfate**	**Phosphate**
Site one	95.63	0.13	34.09	0.01
	72.39	0.43	32.08	0.06
	81.50	0.51	34.52	0.07
	73.88	0.27	31.91	0.00
	73.77	0.42	31.46	0.01
Average	79.434	0.35	32.81	0.03
Standard deviation	±9.738	±0.152823254	±1.387912781	±0.032707981

	Amount (mg/L)			
	Chloride	**Nitrate**	**Sulfate**	**Phosphate**
Site two	72.86	0.56	32.12	0.03
	95.53	0.16	34.19	0.00
	95.67	0.10	34.55	0.00
	95.43	0.08	34.45	0.00
	94.26	0.07	34.34	0.00
Average	90.75	0.19	33.93	0.01
Standard deviation	±10.01396374	±0.206439902	±1.021283162	±0.013483583

	Amount (mg/L)			
	Chloride	**Nitrate**	**Sulfate**	**Phosphate**
Overall Average	85.09181	0.27194	33.37237	0.020666667
Standard deviation	±11.05847665	±0.191143019	±1.290964341	±0.02678955

APPENDIX G: UNFILTERED WATER SAMPLES RAW DATA

	Conductivity (µS/cm)	pH	Alkalinity (mg/l)	Nitrate concentration (mg/l)	phosphate (mg/l)	Dissolved oxygen (mg/l)	Temperature (0^c)
Site one	532	7.42	99	0	0	4.71	15
	524	6.34	98	0	3	3.63	12.1
	580	7.47	120	5	0	3.41	11.4
	554	6.85	123	2	0	3.37	11.7
	500	7.47	156	0	3	3.42	13.3
Mean	538	7.11	119.2	1.4	1.2	3.708	12.7
Standard deviation	±30.39736831	±0.50393452	±23.59449088	±2.19089023	±1.643167673	±0.569227547	±1.474788

	Conductivity	pH	Alkalinity (mg/l)	Nitrate concentration (mg/l)	phosphate (mg/l)	Dissolved oxygen (mg/l)	Temperature (0^c)
Site two	601	8.62	150	0	0	11.73	13.7
	592	7.7	143	0	3	11.03	15.6
	576	7.85	135	1	0	10.74	14.1
	590	8.51	126	0	0	11.6	14.7
	599	8.71	150	0	0	11.6	15
Mean	591.6	8.278	140.8	0.2	0.6	11.34	14.62
Standard deviation	±9.864076237	±0.467621642	±10.32956921	±0.447213595	±1.341640786	±0.431103236	±0.746324

APPENDIX H- TRACE METALS RAW DATA

	Amount (mg/L)				
	Copper	**Zinc**	**Lead**	**Aluminum**	**Nitrite**
Site one	0.000448963	6.32596E-06	0.000363064	0.003356659	0.15
	0.004727662	0.000778944	0.00063418	0.007749029	0
	0.004903206	0.000932721	0.000170236	0.008550613	0.3
	0.004792211	0.001695854	0.000102971	0.010200732	0.15
	0.004637151	0.002231206	0.000190115	0.010323466	0
Mean	0.003901839	0.00112901	0.000292113	0.0080361	0.12
Standard deviation	±0.001932648	±0.00085989	±0.000213937	±0.002835576	±0.125499004

	Amount (mg/L)				
	Copper	**Zinc**	**Lead**	**Aluminum**	**Nitrite**
Site two	0.004580392	0.001201523	0.000403868	0.007682291	0
	0.004538653	0.000708311	3.03848E-05	0.006006299	0
	0.004438818	0.001215497	0.000379566	0.006014586	0
	0.00452839	0.001407089	0.000489198	0.006822762	0
	0.004840459	0.003210007	0.000102971	0.007575675	0
Mean	0.004585342	0.001548485	0.000281197	0.006820323	0
Standard deviation	±0.000151659	±0.00096408	±0.000201656	±0.000810154	±0

	Amount (mg/l)				
	Copper	**Zinc**	**Lead**	**Aluminum**	**Nitrite**
Combined Mean	0.004243591	0.001338748	0.000286655	0.007428211	0.9
Standard deviation	±0.00134166	±0.000889153	±0.000196082	±0.002067813	±0.1048808850

APPENDIX I- DIATOM RAW DATA (DOWNLOADED ON 14/11/2018)

	Genus & Species	Abundance
Lucy H (4B)	Cyclotella meneghiniana	4
	Amphora veneta	2
	Surirella ovalis	2
	Mastogloia smithii	1
	Fragilaria leptostauron var. dubia	2
	Cymbella cistula	2
	Cavinula cocconeiformis	2
	Rhoicosphenia curvata	6
Sivan (10B)	Denticula tenuis	3
	Fragilaria brevistriata	30
	Navicula tuscula	10
	Amphora veneta	2
	Rhoicosphenia curvata	1
	Eunotia valida	3
	Epithemia sorex	6
	Tetracyclus lacustris	1
Ashley	Amphiprora paludosa	1
	Cocconeis pediculus	12
	Fragilaria brevistriata	4
	Navicula menisculus	2
	Cyclotella meneghiniana	3
	Peronia heribaudi	7
	Eunotia exigua	4
	Fragilaria construens	2
	Denticula tenuis	3
	Neidium dubium	1
	Rhoicosphenia curvata	1
	Amphora ovalis	5
Ruddy (A-16)	Navicula digitoradiata	4
	Nitzschia recta	1
	Diatoma vulgaris	1
	Cyclotella meneghiniana	3
	Navicula hodgeana	1
	Luticola sp.	1
Olivia (13A)	Surirella capronii	5
	Cavinula Pseudoscutiform	2
	Amphicampa hemicyclus	30
	Amphora Ovalis	4
	Rhoicosphenia curvata	10
	Cymbella cistula	1
	Fragilaria construens	8

Noah	Navicula tuscula	12
	Fragilaria brevistriata	4
	Eunotia valida	2
	Diatomella balfouriana	1
	Steuroneis phoenicenteron	2
	Surirella Biseriata	1
Amelia	Nupela subrostrata	2
	Cyclotella meneghiniana	4
	Cocconeis pediculus	7
	Fragilaria leptostauron	3
	Amphora veneta	10
	Pinnularia borealis	3
	Cocconeis pseudothumensis	7
	Fragilaria brevistriata	3
	Rhoicosphenia curvata	12
Lily (5B)	Cyclotella kuetizingiana	4
	Diploneis chersononensis	2
	Cocconeis arroniensis	3
	Eunotia valida	1
	Amphora ovalis	2
	Achnanthes bioretii	9
	Pinnularia borealis	32
Joe	Surirella camplodiscus	26
	Fragilaria synedra aslerionella	5
	Aulacoseira eurosolenia	2
	Cyclotella stephanodiscus cyclostephanos	6
	Fragilaria Vaucheriae	2
	Cymbella ventricosa	10
	Amphora veneta	1
Sophie N	Frustulia rhomboides	7
	Navicula farta	7
	Rhoicosphenia curvata	6
	Navicula exigua	13
	Cyclotella comta	8
	Opephora martyi	4
Beth (A7)	Cyclotella comta	4
	Rhoicosphenia curvata	4
	Navicula farta	5
	Achnanthes lanceolata	1
	Gomphonema augur	2
	Opephora martyi	6
	Amphora veneta	1
	Frustulia rhomboides	1

Tom S	Cyclotella comta	2
	Navicula cocconeiformis	1
	Achnanthes lanceolata	2
	Gomphonema parvulum	1
	Stauroneis prominula	1
	Amphipleura pellucida	2
	Cocconeis pediculus	3
	Amphiprora paludosa	4
	Cyclotella meneghiniana	1
	Cymbella ventricosa	2
	Opephora martyi	10
	Nitzschia linearis	1
	Epithemia sorex	1
	Campylodiscus noricus	3
	Hantzschia amphioxys	1
	Synedra ulna	1
	Cocconeis scutellum	1
Hannah Hepworth	Mastogloia Smithii	30
	Diploneis oculata	9
	C. amphisbaena	10
	Diploneis ovalis	1
	Tetracyclus lacustris	1
	Navicula slesvicensis	7
	Amphora veneta	2
	Campylodiscus	1
	Pinnularia stauroptera	1
	Eunotia diodon	15
	Amphora ovalis	3
Bernardine	Thalassiosira fluviatilis	1
	Cyclotella comta	1
	Surirella ovata	1
	Nitzschia linearis	1
	Achnanthes lanceolata	7
	Eunotia valida	8
	Surirella pinnata	1
	Amphora veneta	1
	Cymbella hybrida	9
	Cymbella ventricosa	1
Olivia W	Cyclotella comta	2
	Cymbella ventricosa	3
	Fragilaria vaucheriae	1
	Planothidium lanceolatum	10
	Diatoma vulgaris	3
Alejandra	Nitzschia linearis	2
	Cyclotella meneghiniana	2
	Amphora veneta	4
	Gyrosigma acuminatum	1
	Navicula hodgeana	32
	Fragilaria brevistriata	3
	Stephanodiscus oregonicus	1

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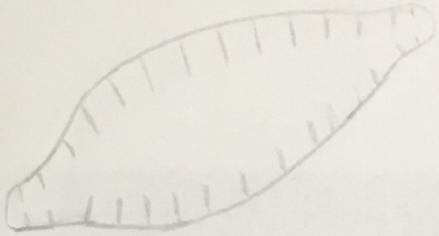
Nathan	Frustulia vulgaris	4
	Navicula crytocephala	27
	Denticula kuetzingii	3
	Opephora olsenii	1
	Mastogloia smithii	5
	Navicula hodgeana	6
	Eunotia rhomboidea	3
	Nitzschia linearis	2
Lu (12B)	Cyclotella meneghiniana	2
	Eunotia lapponica	1
	Fragilaria vaucheriae	2
	Surirella brebissonii	2
Fin(2B)	Cocconeis pediculus	2
	Navicula tuscula	4
	Nitzschia linearis	2
	Amphipleura pellucida	3
	Stephanodiscus astraea	1
	Cyclotella comta	1
	Diatoma hiemale	3
	Navicula menisculus	2
	Amphora ovalis	3
	Diatoma vulgaris	16
Xiaoyi	Cyclotella stephanodiscus cyclostephanos	1
	Surirella camplodiscus	21
	Diploneis ovalis	6
	Pinnularia legumen Ehrenberg	12
	Ovalis Kützing	5
	Cymbella hybrida	6
	N.explanata Hustedt	4
	Gomphonema augur	2
Xi	Cyclotella meneghiniana	5
	Fragilaria brevistriata	4
	Amphora veneta	3
	Amphora ovalis	3
Charlotte	Cyclotella Stephanodiscus Cyclostephanos	6
	Fragilaria brevistriata	4
	Navicula farta	8
	Cymbella ventricosa	7
	Mastogloia smithii	1
	Meridion circulare	5
	(chain of) Fragilaria	4
	Amphora veneta	1
	Navicula cocconeiformis	2
	Pinnularia divergentissima	1
Duncan (C6)	Fragilaria brevistriata	20
	Surirella ovalis	2
	Amphora ovalis	4
	Denticula kuetzingii	12
	Opephora olsenii	1
	Cymbella hybrida	3
	Navicula tuscula	1
	Cyclotella Meneghiniana	3

Chang(C9)	Ovalis Kützing	3
	Rhoicosphenia curvata	1
	Surirella biseriata	2
	Eunotia valida	8
	Navicula pinnularia	4
Z Gu	Cyclotella meneghiniana	3
	Navicula pinnularia	6
R Hanson (K1)	Actinocyclus normanii	1
	Amphicampa hemicyclus	1
	Diploneis ovalis	2
	Fragilaria brevistriata	25
	Surirella camplodiscus	30
Sophie L	Fragilaria brevistriata	7
	Cyclotella stephanodiscus	8
	Denticula kuetzingii	9
	Eunotia faba	3
	Fragilaria construens var. venter	8
	Navicula farta	7
	Diatoma hiemale	5
Joe Gallon	Stephanodiscus	2
	Fragilaria leptostauron	2
	Fragilaria brevistriata	1
	Navicula farta	1
	Navicula exigua	1
	Gyrosigma kützingii	1
Oliver Bevilacqua	Cymbella ventricosa	1
	Amphora veneta	3
	Amphora ovalis	2
	Surirella biseriata	1
	Caloneis latiuscula	1
Nisha	Navicula exigua	2
	Achnanthes lanceolata	1
	Navicula Farta	10
	Denticula tenuis	3
	Stauroneis phoenicenteron	10
	Cocconeis pediculus	1
Ollie (16-c)	Actinocyclus normanii	1
	Surirella Camplodiscus	14
	Eunotia valida	3
George (17, C)	Cyclotella meneghiniana	19
	Cyclotella Stephanodiscus	
	Cyclostephanos	6
	Navicula Farta	15
	Surirella Camplodiscus	13
Laura	Nitzschia Denticula	9
	Navicula Meniscus	25
	Bacilleria Genera	1
	Amphipleura pellucida	10
	Stephanodiscus Astraea	4
	Nitzschia linearis	1
	Diatoma Hiemale	1

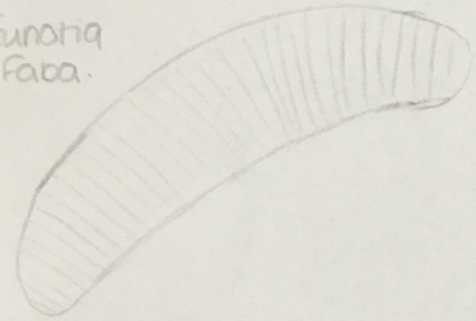
Phytobenthos identification

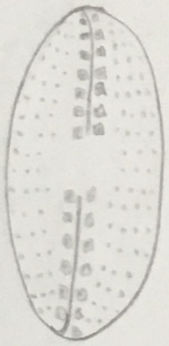
Fragilaria brevistriata



Diatoma hiemale

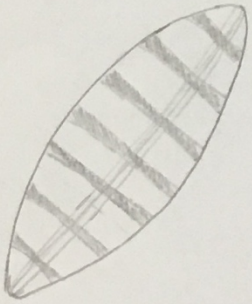
Eunotia Faba.



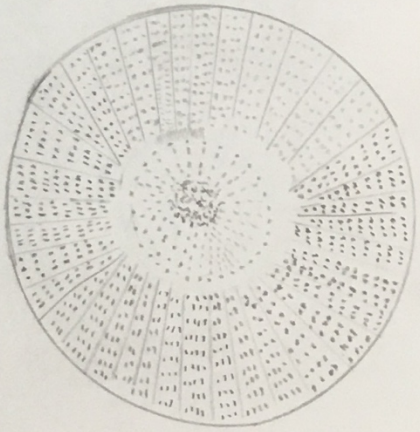


Navicula Farta.

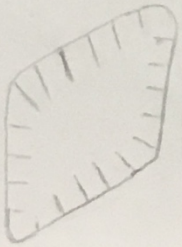
Denticula Kützing



Stephanodiscus Astraea.



Fragilana construens var. venter



APPENIDX J- WORKING CALCULATIONS FOR DIATOM EQR

Genus & Species	Total abundance (a)	Nutrient sensitivity score (s)	a x s
Achnanthes bioretii	9	3	27
Achnanthes lanceolata	11	5	55
Actinocyclus normanii	1	0	0
Amphicampa hemicyclus	30	0	0
Amphiprora paludosa	5	0	0
Amphipleura pellucida	5	2	10
Amphora ovalis	26	5	130
Amphora veneta	28	5	140
C. amphisbaena	10	0	0
Caloneis latiuscula	1	0	0
Campylodiscus	1	0	0
Campylodiscus noricus	3	0	0
Cavinula cocconeiformis	2	3	6
Cavinula Pseudoscutiform	2	3	6
Cocconeis arroniensis	3	0	0
Cocconeis pediculus	25	5	125
Cocconeis pseudothumensis	7	3	21
Cocconeis scutellum	1	0	0
Cyclotella comta	18	0	0
Cyclotella kuetizingiana	4	0	0
Cyclotella meneghiniana	30	0	0
Cymbella cistula	3	3	9
Cymbella hybrida	12	0	0
Cymbella ventricosa	7	0	0
Denticula kuetzingii	15	4	60
Denticula tenuis	9	2	18
Diatoma hiemale	3	0	0
Diatoma vulgaris	20	5	100
Diatomella balfouriana	1	0	0
Diploneis chersononensis	2	0	0
Diploneis oculata	9	4	36
Diploneis ovalis	1	3	3
Epithemia sorex	7	3	21
Eunotia diodon	15	1	15
Eunotia exigua	4	1	4
Eunotia lapponica	1	0	0
Eunotia rhomboidea	3	1	3
Eunotia valida	22	1	22
Fragilaria brevistriata	69	4	276
Fragilaria construens	10	4	40
Fragilaria leptostauron	5	5	25
Fragilaria leptostauron var. dubia	2	5	10
Fragilaria vaucheriae	3	4	12
Frustulia rhomboides	8	1	8
Frustulia vulgaris	4	1	4
Gomphonema augur	2	5	10
Gomphonema parvulum	1	5	5
Gyrosigma acuminatum	1	5	5
Gyrosigma kützingii	1	0	0
Hantzschia amphioxys	1	4	4
Luticola sp.	1	3	3
Mastogloia smithii	36	3	108
Navicula cocconeiformis	1	3	3
Navicula crytocephala	27	4	108

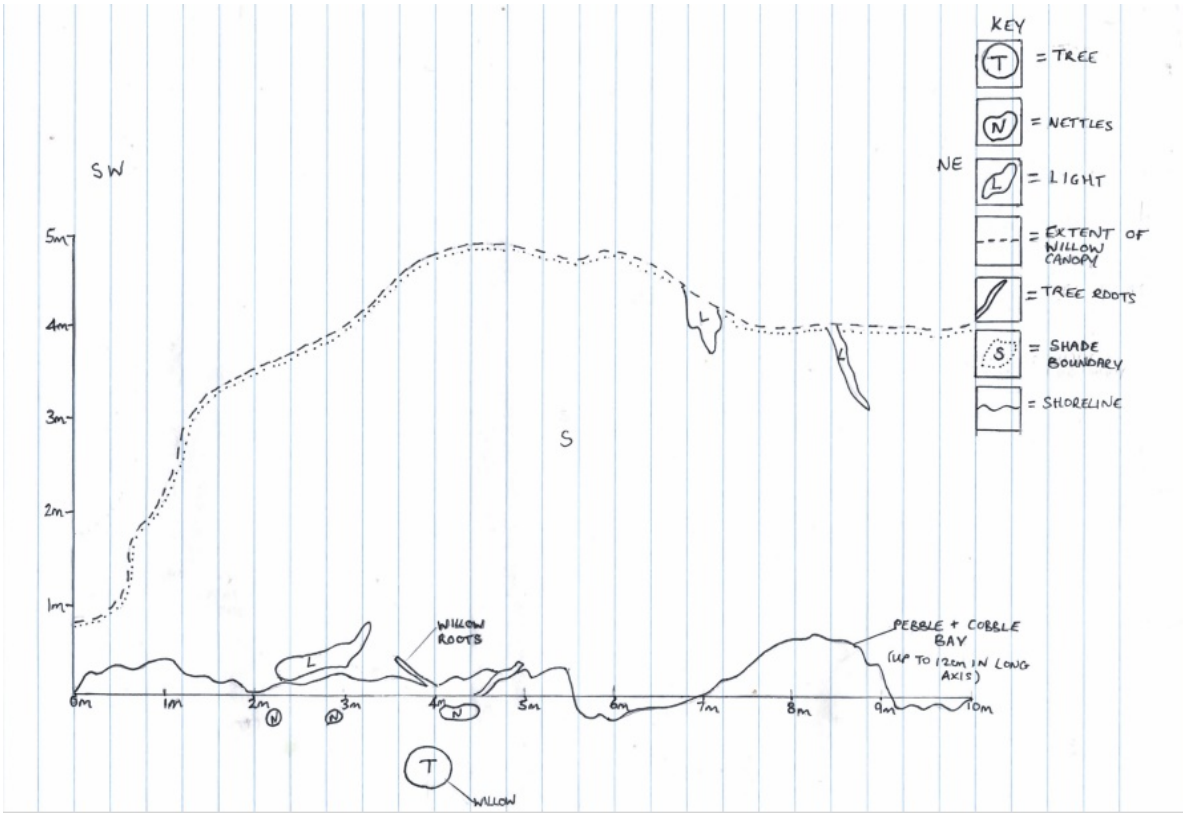
Navicula digitoradiata	4	4	16
Navicula exigua	16	0	0
Navicula Farta	23	0	0
Navicula hodgeana	39	0	0
Navicula menisculus	4	5	20
Navicula pinnularia	10	0	0
Navicula slesvicensis	7	4	28
Navicula tuscula	27	3	81
Neidium dubium	1	0	0
Nitzschia linearis	8	4	32
Nitzschia recta	1	4	4
Nupela subrostrata	2	0	0
Opephora martyi	20	0	0
Opephora olsenii	2	4	8
Ovalis Kützing	3	0	0
Peronia heribaudi	7	0	0
Pinnularia borealis	35	2	70
Pinnularia stauroptera	1	0	0
Planothidium lanceolatum	10	5	50
Rhoicosphenia curvata	41	5	205
Stauroneis phoenicenteron	10	0	0
Stauroneis prominula	1	0	0
Stephanodiscus astraea	1	0	0
Stephanodiscus oregonicus	1	0	0
Steuroneis phoenicenteron	2	0	0
Surirella Biseriata	4	3	12
Surirella brebissonii	2	4	8
Surirella capronii	5	0	0
Surirella ovalis	4	3	12
Surirella ovata	1	0	0
Surirella pinnata	1	0	0
Synedra ulna	1	3	3
Tetracyclus lacustris	2	1	2
Thalassiosira fluviatilis	1	0	0

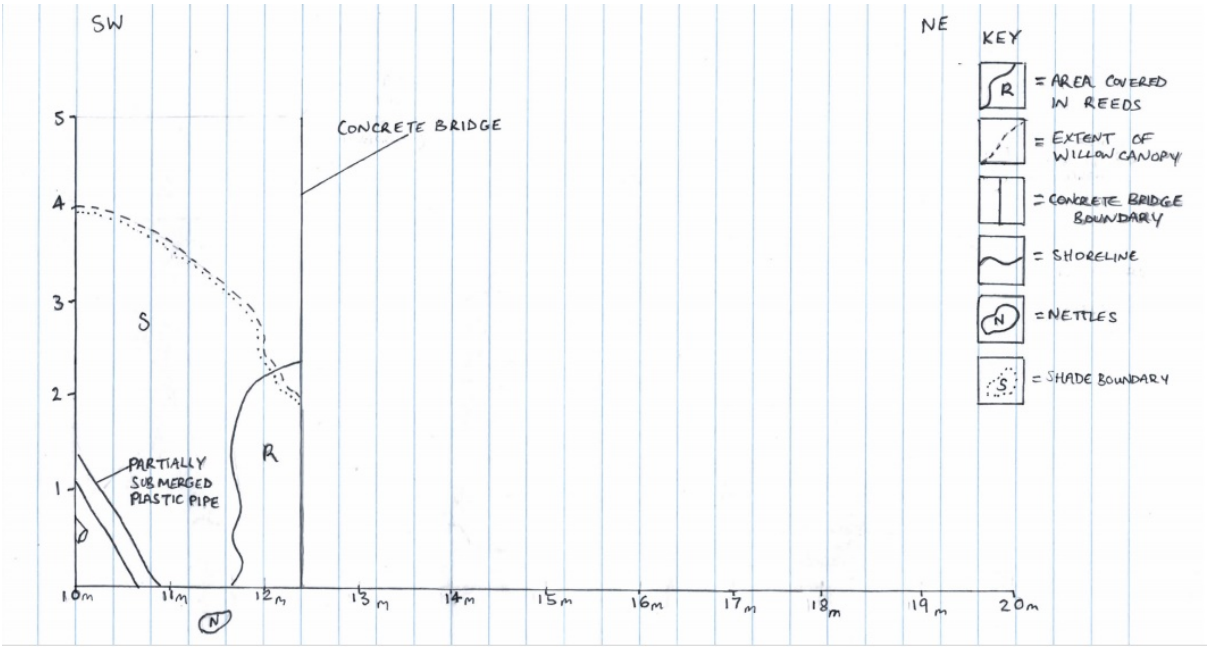
Sum of a =	825		
Sum of as =			1983
Calculate w = sum as/sum a =	2.403636364		
(W x 25) - 25 =			
(2.40363636363636x25)-25=	35.09090909		
Reference LTDI2 value (from alkalinity)	35		
EQR =			
(100 - observed LTDI2) / (100 - reference LTDI2)			
(100-35.09)/(100-35)	0.99861538		
If EQR is greater than 1, set to 1	High?		

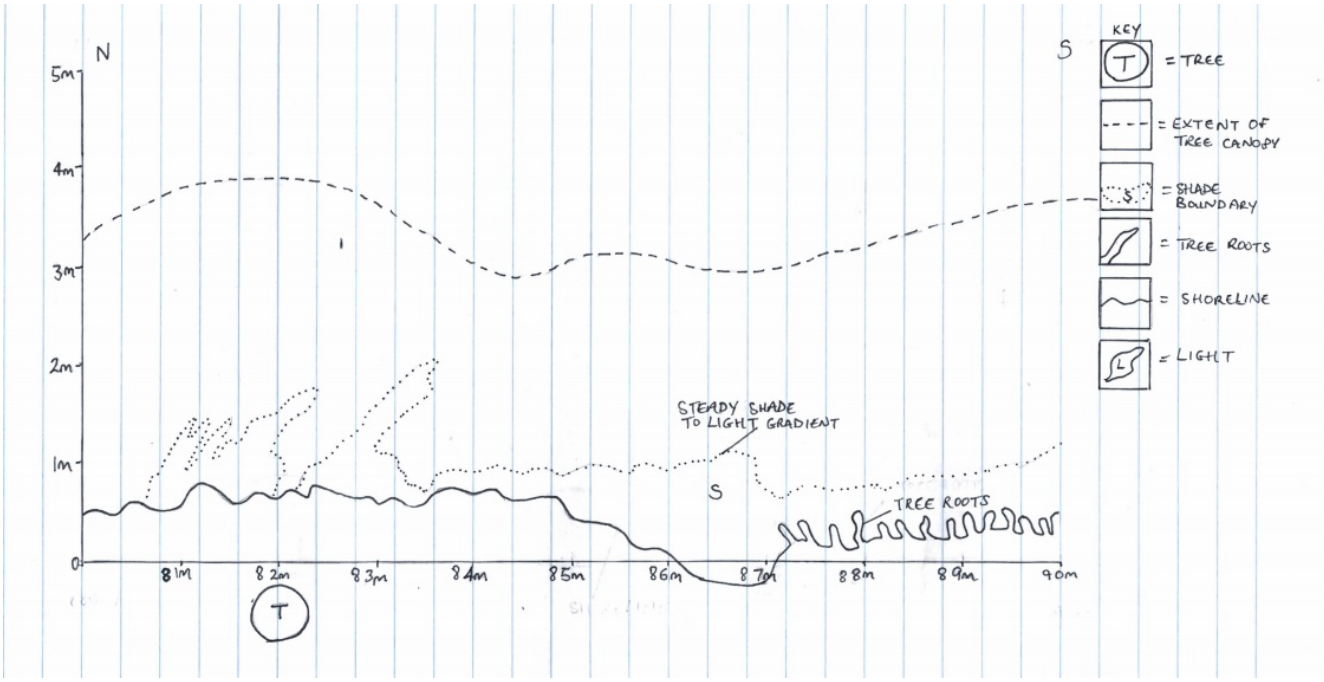
APPENDIX K-MACROPHYTE COVER

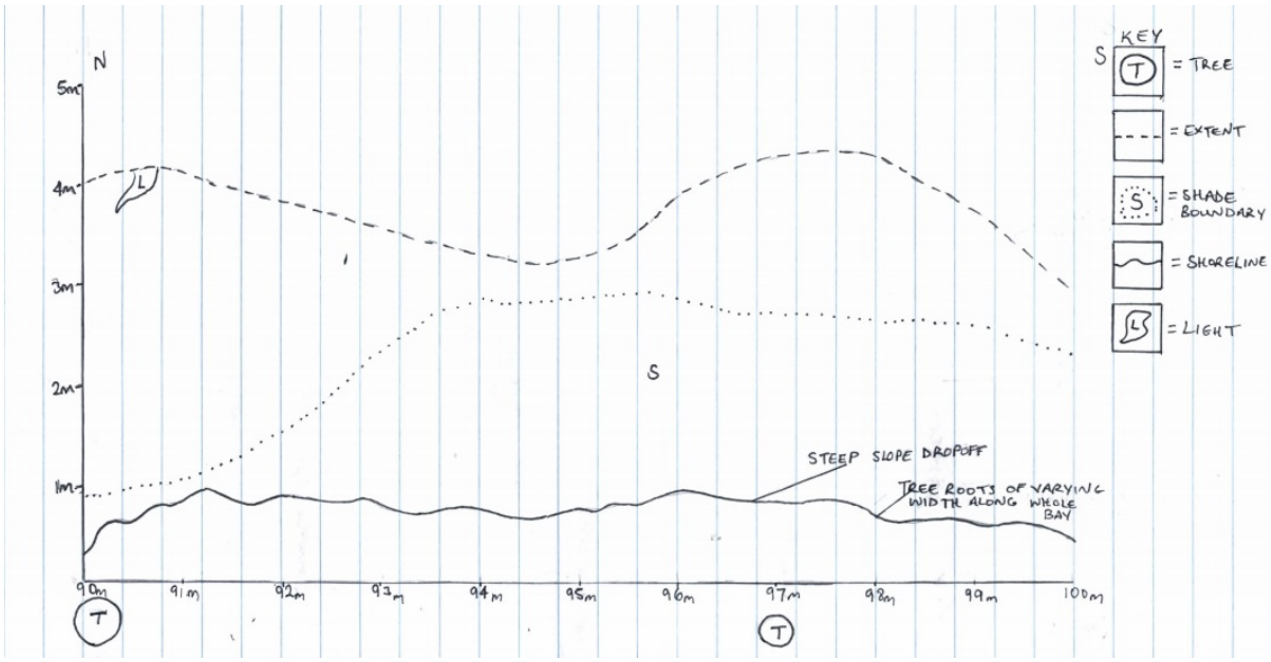
Site 1. Starting point at 80 [m] Reference points	Reed	% Cover	Length of Bank (m)
0			
2			
4			
6			
8			
10			
12			
14			
16			
18			
20	Phragmites australis	70	1.4
22	Phragmites australis	80	1.6
24	Phragmites australis	60	1.2
26	Phragmites australis	30	0.6
28	Phragmites australis	5	0.1
30	Phragmites australis	0	0
32			
34			
36			
38			
40			
42			
44			
46			
48	reeds		
50	reeds		
52	reeds		
54	reeds		
56	reeds		
58	reeds		
60			
62	*Glyceria maxima*	45	0.9
64			
66			
68			
70			
72			
74			
76			
78			
80			
82			
84			
86			
88			
90			
92	Bridge	0	0
94	Bridge	0	0
96	Bridge	0	0
98	Bridge	0	0
100	Bridge	0	0

APPENDIX L- SITE SKETCHES






N
S
5m
4m
3m
2m
1m
0
KEY
= TREE
= EXTENT OF TREE CANOPY
= SHADE BOUNDARY
= TREE ROOTS
= SHORELINE
= LIGHT
STEADY SHADE TO LIGHT GRADIENT
TREE ROOTS
S
81m
82m
83m
84m
85m
86m
87m
88m
89m
90m
T


N
S
5m
4m
3m
2m
1m
KEY
= TREE
= EXTENT
= SHADE BOUNDARY
= SHORELINE
= LIGHT
L
S
STEEP SLOPE DROPOFF
TREE ROOTS OF VARYING WIDTH ALONG WHOLE BAY
90m
91m
92m
93m
94m
95m
96m
97m
98m
99m
100m
T
T