MEMORANDUM

Bioresearches
A Babbage Company

TO:	Watercare services Ltd	Date:	2 June 2025
COPY TO:	Diana Villa Ortega	Job No:	68023
FROM:	Laureline Meynier		

WAIUKU WASTEWATER TREATMENT PLANT – MONITORING OF OYSTER QUALITY 2025

The Waiuku wastewater treatment plant requires annual environmental monitoring for their operations in the Waiuku estuary under discharge permit DIS60334129. Watercare Services Ltd contracted Bioresearches to conduct oyster quality monitoring during the 2024-2025 summer period.

Specified in consent conditions 15 to 17, Pacific oysters shall be collected annually in February from five monitoring sites.

This memorandum presents the results from the annual oyster quality monitoring in February 2025.

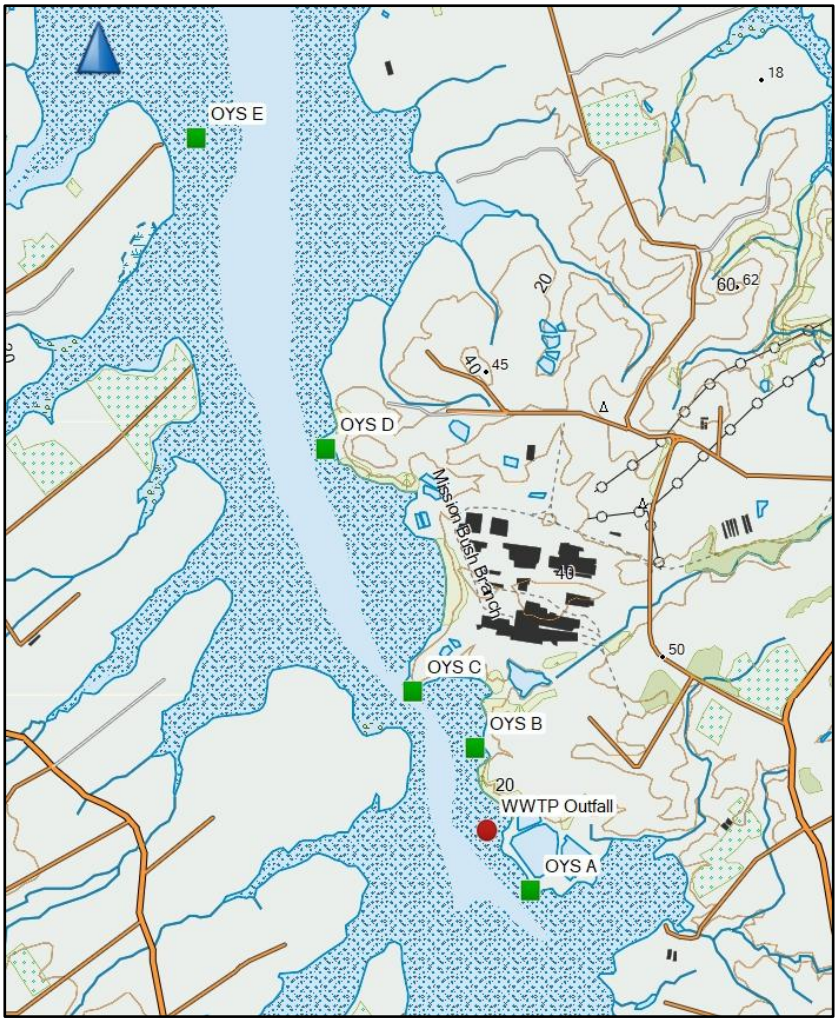

Figure 1 Monitoring Sites for oyster quality

Bioresearches
Level 4, 68 Beach Road, Auckland 1010
PO Box 2027, Shortland Street,
Auckland 1140, New Zealand
T 09 379 9980 **F** 09 377 1170
E admin@babbage.co.nz **www.bioresearches.co.nz**

Oyster sampling methodology

Following the consent conditions 15 to 17, five replicates of 12 oysters (60 to 70 mm) were sampled at each of the five monitoring sites described below (Figure 1):

- **OYS A**: a site upstream of the discharge point, with a minimum separation distance of 250m
- **OYS B**: a site approximately half the distance from the discharge point and the Needles site
- **OYS C**: a site adjacent to the Needles
- **OYS D**: a site adjacent to Okohaka Point
- **OYS E**: a site adjacent to Gordon's Landing

The weather conditions on the day of sampling and the week prior sampling were noted, as well as the tide characteristics and time of sampling. The time of discharge release, the discharge volume, rate and quality were not provided here. The oyster results were not interpreted in relation to the discharge data as it was out of scope of the memorandum. After collection, oysters were returned to the laboratory at Bioresearches, cleaned, and removed from shells into sterile jars, then delivered chilled to the Watercare Services Laboratory on the same day.

Oysters were analysed for Escherichia coli by MPN (Most Probable Number) as consent condition 17. Faecal coliform numbers were used historically for shellfish quality assessment but have been substituted in recent years by E. coli testing to align with European guidelines (NZFSA, 2006). To keep a dataset consistent with previous years, faecal coliforms were still tested along E. coli. The methodology was based on the APHA (American Public Health Association) 9221. The guidelines for faecal coliforms (Ministry of Health, 1995) and for E. coli (NZFSA, 2006) are presented in

Table 1.

Table 1. Guidelines for bacterial indicators in shellfish (MPN) for New Zealand

Test	Median MPN limit / 100g (for 5 replicates)	Max MPN limit / 100g	References
Escherichia coli	230	No more than 10% > 700	NZFSA, Ministry for Primary Industries, 2006
Faecal coliforms	no more than 2 samples out of 5 > 230	No sample > 330	Ministry of Health guidelines, 1995

Results and conclusion

The oysters were collected on the 24th of February 2025 around low tide. The approximate time of sampling for each of the five oyster sites is displayed in Table 2. In the week prior to sampling, the weather

was sunny with no rain being recorded. On the day of sampling, weather conditions were sunny with a light wind.

The MPN of *Escherichia coli* and faecal coliforms are presented in Table 3 and Table 4 respectively. The certificate of analysis by Watercare Laboratory Services is presented in Appendix A.

Table 2 Time of oyster sampling for each site, 24 February 2025

Station	Time of Sampling	Time of low tide
OYS A (250m Upstream)	13:50 – 14:00	14:45 (Onehunga)
OYS B (250m Downstream)	14:05 – 14:15	
OYS C (Needles)	14:25 – 14:35	
OYS D (Okohaka Point)	14:40 – 14:50	
OYS E (Gordons Landing)	14:55 – 15:05	

Table 3 Escherichia coli numbers in shellfish samples (MPN/100g wet weight)

	Replicates					
Station	**1**	**2**	**3**	**4**	**5**	**Median**
OYS A (250m Upstream)	<18	<18	<18	<18	<18	<18
OYS B (250m Downstream)	330	230	130	330	45	230
OYS C (Needles)	<18	330	130	45	78	78
OYS D (Okohaka Point)	<18	<18	<18	<18	<18	<18
OYS E (Gordons Landing)	<18	<18	<18	20	<18	20

The oyster tissues collected in February 2025 showed levels of *E coli* and faecal coliforms below the guidelines, except for site OYS B 250m downstream of the outfall (Tables 3 and 4). OYS C site, further downstream, gave significant numbers of bacteria but under the guideline values. OYS B site was therefore not compliant with shellfish standards. The downstream sites D and E, and the upstream site A showed little to no bacteria in the oyster tissues.

The large numbers of faecal bacteria in OYS B site may be influenced by the Waiuku wastewater treatment plant effluent. However, the inspection of the previous annual survey results together with the weather data preceding the sampling, revealed that several factors contribute to the source of faecal bacterial loads in the Waiuku coastal waters, and subsequently in oysters (Figure 2).

Table 4 ***Faecal coliform numbers in shellfish samples (MPN/100g wet weight)***

	Replicates					
Station	**1**	**2**	**3**	**4**	**5**	**Median**
OYS A (250m Upstream)	<18	20	<18	<18	<18	20
OYS B (250m Downstream)	330	230	130	490	78	230
OYS C (Needles)	<18	330	130	45	130	130
OYS D (Okohaka Point)	<18	<18	<18	<18	<18	<18
OYS E (Gordons Landing)	<18	<18	<18	20	<18	20

The E. coli presence in oyster tissue over the 5 sites during the last two surveys (February 2024 and 2025) showed a similar profile with the highest concentration at OYS B, less at OYS C, and close to inexistant at the other sites (Figure 2). The annual surveys in 2024 and 2025 were conducted after at least 7 days of dry weather, when run-off with possible diffuse pollution was minimal. Prior to 2024, the sampling was conducted after significant rain events and high bacteria loads were present not only in oysters at sites B and C, but also in oysters further away and upstream. Specifically, the site OYS E at Gordons Landing (farmland west side of the catchment) showed very high numbers of E. coli in oysters during the 2020 to 2022 surveys after rain events (Figure 2).

The amount of rain during the week preceding sampling has significant effects on the concentrations of contaminants, including bacteria, on waterways. The Waiuku catchment consists mostly of farmland, especially on its western side. The eastern part of the catchment has an industrial site (New Zealand Steel Ltd) and expanding residential areas. Faecal bacteria in the oysters around the outfall are therefore likely to originate from multiple pollution sources (wastewater discharge, farmland animals, birds) exacerbated by rain events with the increase of run-offs.

The use of genetic markers on water samples close to the oyster sites would allow the identification of the source of faecal bacteria in oysters. Indeed, microbial source tracking is now a common approach for faecal source identification in aquatic environments (Paruch and Paruch, 2022; Muirhead *et al.,* 2023).

Regards,

Laureline Meynier, Marine Ecologist
Bioresearches
A Babbage Company
Babbage Consultants Limited
Level 4, 68 Beach Road, Auckland 1010
PO Box 2027, Shortland Street, Auckland 1140, New Zealand
M 021 123 7865 **E** Laureline.meynier@bioresearches.co.nz

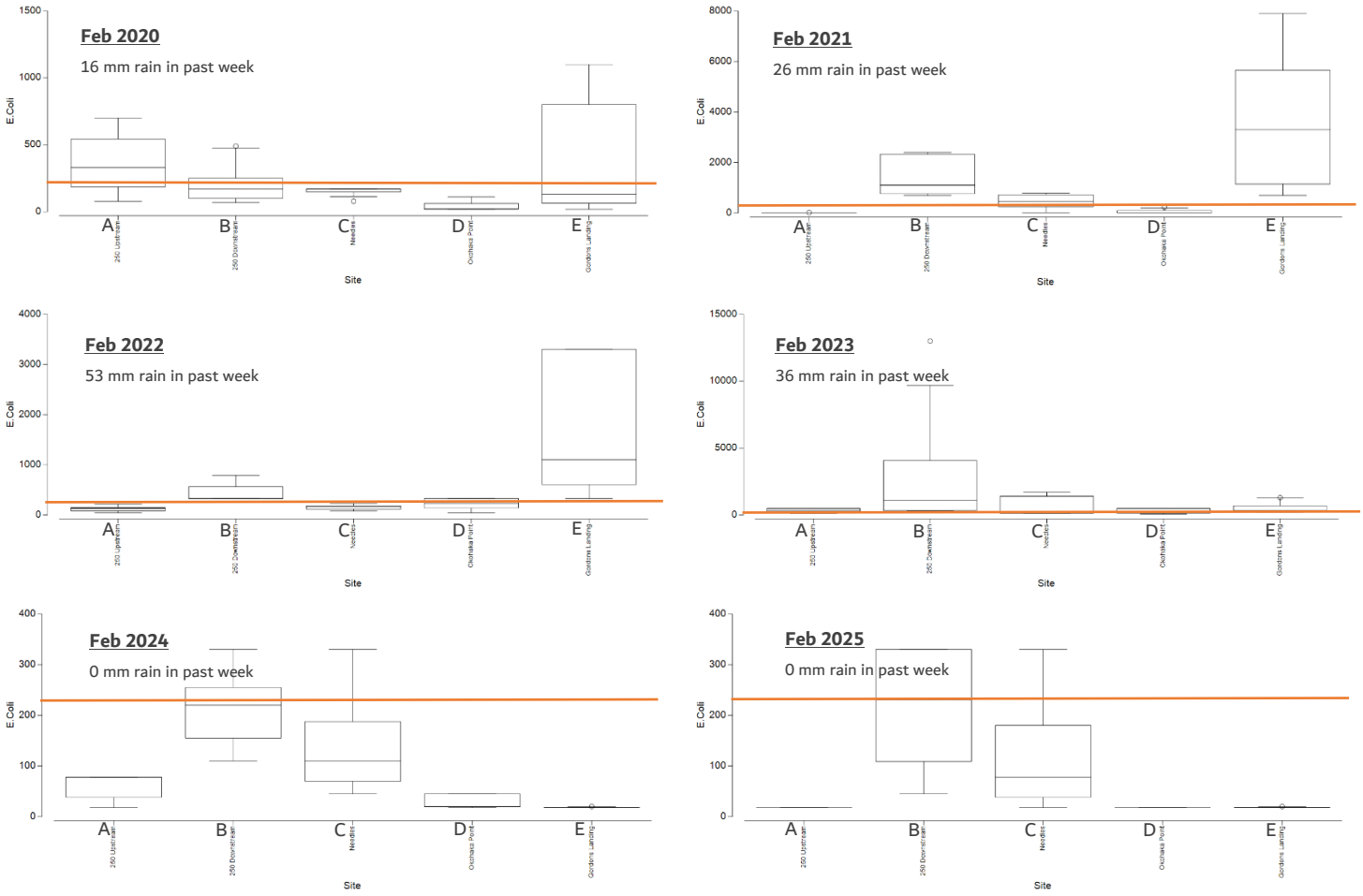

Figure 2 E. coli MPN at each oyster site during the annual surveys since 2020. The orange line represents the guidelines limit.

References

Ministry of Health (1995)

Microbial reference Criteria for Food, Ministry of Health, Wellington.

NZFSA (2006)

New Zealand Food Safety Authority. Animal products specifications for bivalve molluscan shellfish. https://www.mpi.govt.nz/processing/seafood/seafood-processing/bivalve-molluscan-shellfish-bms/growers-and-harvesters-of-bivalve-molluscan-shellfish-bms.

Muirhead R. Hudson R., Cookson A. (2023)

A review of river microbial water quality data in Northland region. Prepared by AgResearch for the Northland Regional Council

Parush and Parush (2022)

An overview of microbial source tracking using host-specific genetic markers to identify origins of fecal contamination in different water environments. Water 14: 1809. https://doi.org/10.3390/w14111809.

APPENDIX A

Oysters certificate of Analysis – 24 February 2025

Watercare
Laboratory Services

Watercare Services Limited
52 Aintree Ave, Māngere, Auckland, 2022
PO Box 107028, Auckland, 2150
T: 0800 522 365
clientsupport@water.co.nz
www.watercarelabs.co.nz

Certificate of Analysis
Laboratory Reference: 250222-038

Attention:	Laureline Meynier	*Final Report:*	**588329-0**	*Replaces Report*	**581090-0**
Client:	**WATERCARE SERVICES LTD**	*Report Issue Date:*	**02-May-2025**		
Address:	**500 Island Road, Mangere, Auckland, 2022**	*Received Date:*	**25-Feb-2025**		
		Sampled By:	**LM**		
Client Reference:	**Waiuku Wastewater Treatment Plant - Shellfish Quality**	*Laboratory Activity Dates:*	**25-Feb-2025** -	**01-Mar-2025**	
Purchase Order:	**3310-OP11-693040**	*Quote Reference :*	**16308**		

Amended CoA generated with corrected accreditation status.

Sample Details					
Lab Sample ID:		**250222-038-1**	**250222-038-2**	**250222-038-3**	**250222-038-4**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-UP250 (A) 1	OYS-UP250 (A) 2	OYS-UP250 (A) 3	OYS-UP250 (A) 4
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	<18	<18	<18	<18
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	<18	20	<18	<18

Sample Details					
Lab Sample ID:		**250222-038-5**	**250222-038-6**	**250222-038-7**	**250222-038-8**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-UP250 (A) 5	OYS-DS250 (B) 1	OYS-DS250 (B) 2	OYS-DS250 (B) 3
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	<18	330	230	130
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	<18	330	230	130

Sample Details					
Lab Sample ID:		**250222-038-9**	**250222-038-10**	**250222-038-11**	**250222-038-12**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-DS250 (B) 4	OYS-DS250 (B) 5	OYS-Needles (C) 1	OYS-Needles (C) 2
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	330	45	<18	330
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	490	78	<18	330

Sample Details					
Lab Sample ID:		**250222-038-13**	**250222-038-14**	**250222-038-15**	**250222-038-16**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-Needles (C) 3	OYS-Needles (C) 4	OYS-Needles (C) 5	OYS-Okohaka (D) 1
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	130	45	78	<18
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	130	45	130	<18

Report Number: **588329-0** Watercare Laboratory Services **Page 1 of 3**

Sample Details					
Lab Sample ID:		**250222-038-17**	**250222-038-18**	**250222-038-19**	**250222-038-20**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-Okohaka (D) 2	OYS-Okohaka (D) 3	OYS-Okohaka (D) 4	OYS-Okohaka (D) 5
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	<18	<18	<18	<18
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	<18	<18	<18	<18

Sample Details					
Lab Sample ID:		**250222-038-21**	**250222-038-22**	**250222-038-23**	**250222-038-24**
Client Sample ID:					
Sample Date/Time		24/02/2025	24/02/2025	24/02/2025	24/02/2025
Description:		OYS-Gordon (E) 1	OYS-Gordon (E) 2	OYS-Gordon (E) 3	OYS-Gordon (E) 4
Microbiology					
Escherichia coli by MPN					
Escherichia coli (MPN)	MPN/100g	<18	<18	<18	20
Faecal coliforms by MPN					
Faecal coliforms (MPN)	MPN/100g	<18	<18	<18	20

Sample Details			
Lab Sample ID:		**250222-038-25**	
Client Sample ID:			
Sample Date/Time		24/02/2025	
Description:		OYS-Gordon (E) 5	
Microbiology			
Escherichia coli by MPN			
Escherichia coli (MPN)	MPN/100g	<18	
Faecal coliforms by MPN			
Faecal coliforms (MPN)	MPN/100g	<18	

*Results marked with * are not accredited to International Accreditation New Zealand. A dash indicates no test performed.*

Where samples have been supplied by the client, they are tested as received.

The results of analysis contained in this report relate only to the sample(s) tested. Where sample collection was performed by the laboratory, the results of analysis contained in this report relate only to the sample(s) collected.

Reference Methods

The sample(s) referred to in this report were analysed by the following method(s)

Analyte	Method Reference	MDL	Samples	Location
Microbiology				
Escherichia coli by MPN				
Escherichia coli (MPN)	In-house based on APHA 9221	2 MPN/100g	All	Auckland
Preparations				
Faecal coliforms (MPN)	In-house based on APHA 9221	2 MPN/100g	All	Auckland
Shellfish Blending	In-house method		All	Auckland

The method detection limit (MDL) listed is the limit attainable in a relatively clean matrix. If dilutions are required for analysis the detection limit may be higher. For more information please contact the Compliance and Projects Manager.

Samples, with suitable preservation and stability of analytes, will be held by the laboratory for a period of two weeks after results have been reported, unless otherwise advised by the submitter.

Watercare Laboratory Services is a division of Watercare Services Limited .

Robyn Abernethy
Compliance and Projects Manager