

**UPL Chemical Spill
Toxicity Results following Ultraviolet and Ozone treatment
of PCD Water**

Nicolette T. Forbes

Marine and Estuarine Research (MER), P.O. Box 417, Hyper by the Sea, 4053

TABLE OF CONTENTS

1	PREFACE	1
2	INTRODUCTION	1
3	RESULTS AND DISCUSSION	2
	3.1 ORGANIC PESTICIDE AND METAL LEVELS.....	2
	3.2 TOXICITY AFTER UV / OZONE AND ION-EXCHANGE TREATMENT.	4
4	CONCLUSIONS	5
5	APPENDIX A: CERTIFICATES FROM V&M-ATLS WITH THE PESTICIDE AND METAL RESULTS RESULTS FOR WATER 23 MAY 2023	6

Citation - when using information from this report please reference in full as follows:
Forbes, N.T. (2023). UPL Chemical Spill: Toxicity Results following UV and Ozone treatment of PCD water MER Specialist Report 12/UPL/21.

1 PREFACE

This report is submitted as a stand-alone document focussed on toxicity tests of water collected from the PCD and following UV and Ozone (O₃) treatment and a metal ion exchange column. This version of the report deals only with the results of chemical analysis for metalloid, organic pesticide concentrations and sea urchin toxicity testing of the PCD water pre- and post- the specified treatments in late May 2023.

2 INTRODUCTION

MER, with the assistance of Mr Tex Westgate of WaterScience, sampled water from the PCD as well as water treated with a combination of UV and Ozone and then a further sample of this treated water passed through a metal exchange column. A sample from the tributary was also taken at the same time. All four samples were sent to V&M ATLS for analytical testing of

metals and inorganic compounds and a duplicate sample sent to the CSIR for sea urchin ecotoxicity testing. Results are reported in Section 3.

3 RESULTS AND DISCUSSION

3.1 Organic pesticide and metal levels.

A total of fifty-five pesticide standards were used to analyse the PCD water sample taken on the 23 May 2023 and of these seven were present in the PCD. Four of these were present but below quantifiable levels. Three, viz amicarbazone, metalochlor and tebuthiuron were present at similar quantifiable concentrations (Table 3-1). (Certificates attached in Appendix A).

The PCD water was treated with Alum to flocculate silt and then subjected to the O₃/UV treatment plant which incorporated a final sand and activated charcoal filter at the end point. The results following this treatment show a substantial reduction in the organic pesticides. This confirmed earlier test results using this process¹ (MER 2023) which also indicated a significant improvement in chemical composition of the PCD water following this treatment.

In the above case the remaining organic pesticides were removed almost completely after treatment with the exception of the three compounds mentioned above which remained but in substantially lower concentrations. All three were below the limits of accurate quantification (<30 µg/l). The O₃/UV treated water was then passed through an ion-exchange column to remove metals. This seemed to completely remove the remaining pesticides with the exception of a trace amount of tebuthiuron. This is attributed to the remaining organic pesticides being adsorbed to the ion-exchange column (Verdoorn² *pers comm*).

The results for the removal of metalloids using the ion-exchange column after the O₃-UV treatment were a bit more varied (Table 3-2). Arsenic was reduced slightly by the two treatments used in sequence but was still not brought anywhere close to the background levels in the natural environment which should be in the region of 1-2.6 µg/l. Copper seemed to be reduced by the first treatment through the O₃/UV system and this may be through the final sand filter/activated carbon step. The ion-exchange treatment did not significantly change this value.

¹ Forbes, N.T. (2023). UPL Chemical Spill: Toxicity Results after UV and Ozone treatment applied to PCD water. MER Specialist Report 10/UPL/21.

² Dr G. Verdoorn | Griffon Poison Information Centre telephonic discussions about the results on the 5/6 and 8/6

Manganese showed the greatest response to the ion-exchange treatment following the O₃/UV step. The results indicate a slight reduction in manganese after the first step and substantial reduction after the ion-exchange treatment from 28.3 µg/l in the PCD to 1.3 µg/l.

Zinc results were the most puzzling with more zinc being recorded post-O₃/UV followed by a slight reduction following exposure to the ion exchange column. It is suggested that the final stage on the O₃/UV plant, which comprises a sand/activated charcoal series, may remove the MSMA arsenic fraction but would not remove all elemental arsenic present in solution. This may explain the slight reduction in arsenic concentration in the two-step treatment process as both have activated carbon and fine sand filter layers.

Table 3-1 Organic pesticide concentrations pre- and post-UV and Ozone treatment (all values in µg/l) - note that the water from the PCD (SOURCE) is put through the first treatment (columns 2 results) and then through the ion-exchange column (column3). The tributary (TRIB) is added as a background comparison.

Sample code Compound tested	SOURCE	TWO-STEP TREATMENT		BACKGROUND
	PCD µg/l	POST O ₃ /UV µg/l	POST- ION EXCHANGE µg/l	TRIB µg/l
Ametryn	0	1	0	1
Amicarbazone	240	20	0	9
Azoxystrobin	1	0	0	0
EPTC	3	0	0	0
Metolachlor	216	19	0	7
Tebuconazole	2	0	0	0
Tebuthiuron	234	28	4	19

Table 3-2 Metalloid concentrations in the PCD water, post-UV/Ozone, and following removal of metals through ion-exchange - note that the water from the PCD (SOURCE) is put through the first treatment (columns 2 results) and then through the ion-exchange column (column 3). The tributary (TRIB) is added as a background comparison.

Sample code Metal tested	SOURCE	TWO-STEP TREATMENT		BACKGROUND
	PCD µg/l	POST O ₃ /UV µg/l	POST ION-EXCHANGE µg/l	TRIBUTARY µg/l
Arsenic	6.9	4.3	4.0	1.2
Copper	5.9	2.9	2.8	3.7
Manganese	28.3	17.3	1.3	774.7
Zinc	129	171.7	142.6	98

3.2 Toxicity after UV / Ozone and Ion-exchange Treatment.

The PCD sample taken directly from the dam showed the water to be Marginally Toxic (Table 3-1) and an improvement from the January results (possibly as a result of breakdown time or dilution through the much fuller PCD). After flocculation treatment with alum the raw undiluted sample which was passed through the O₃/UV treatment improved to a Slightly Toxic category. This was a slight decrease in quality from January when the clarified raw water post O₃/UV was rated Not Toxic. The clarified water was then passed through an ion-exchange plant to address the metalloid constituents and analysis produced a Not Toxic result.

Table 3-3 Toxicity results of water using the sea urchin embryo-larval development test in May 2023.

Medium	Treatment	23 May 2023			
		SOURCE	TWO-STEP TREATMENT		BACKGROUND
		PCD direct water sample	POST O ₃ /UV	POST ION-EXCHANGE	TRIBUTARY
WATER	Raw	Marginally Toxic	Slightly Toxic	Not Toxic	Slightly Toxic
	1:5 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:10 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:20 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:40 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:100 dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:200 dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:400 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic

Table 3-4 Toxicity results of water using the sea urchin embryo-larval development test in January 2023 for comparison.

Medium	Treatment	12 January 2023			
		PCD direct water sample	Post-flocculation	Post-UV/O ₃ treatment	Tributary
WATER	Raw	Moderately Toxic	Highly Toxic	Not Toxic	Marginally Toxic
	1:5 Dilution	Not Toxic	Moderately Toxic	Not Toxic	Not Toxic
	1:10 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:20 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:40 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:100 dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:200 dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic
	1:400 Dilution	Not Toxic	Not Toxic	Not Toxic	Not Toxic

Toxicity Category
Inhibition >50% = highly toxic
Inhibition >20 and ≤ 50% = moderately toxic
Inhibition >10% and ≤ 20% = marginally toxic
Inhibition > Minimum Significant Difference % and ≤ 10% = slightly toxic
Inhibition ≤ Minimum Significant Difference% = not toxic

4 CONCLUSIONS

The O₃/UV treatment has produced good results after January 2023 (Not Toxic) and May 2023 (Slightly Toxic). The addition of the ion-exchange column treatment step post-O₃/UV treatment in May 2023 also resulted in the final water being categorised as Not Toxic. It is recommended that during the next round of testing a post-floc sample is tested after the alum treatment to understand the toxicity category of the next step in the treatment. It is MER's understanding that the ion-exchange column will be upgraded to test its efficacy with the metal removal again and this will provide a repeat test of all the treatments.

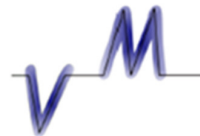
N. Forbes

8 June 2023

5 APPENDIX A: CERTIFICATES FROM V&M-ATLS WITH THE PESTICIDE AND METAL RESULTS RESULTS FOR WATER 23 MAY 2023

V&M Analytical Toxicology Laboratory Services (Pty) Ltd

83 Victoria Street, George, 6529 • PostNet Suite 254, P. Bag X6590, George, 6530
Tel: +27 (0) 44 874 8484/ 873 4153 • Fax: +27 (0) 86 725 8919
Mobile: +27 (0) 82 520 2210/ +27 (0) 82 775 6932
Accurate • Reproducible • Cost effective • **RESULTS** • On time-Every time



CONFIDENTIALITY CAUTION

This message is intended only for the use of the individual or entity to which it is addressed and contains information that is privileged and confidential. If you have received this communication in error, please notify the sender of the e-mail immediately by return e-mail and delete it and any attachment(s) from your system immediately. If you are not the intended recipient, you are also hereby notified that any dissemination, distribution or copying of this communication is strictly prohibited.

TEST REPORT			
Date	05 June 2023	Report No.	230601 VM31489 p
To	Marine & Estuarine Research (MER)	Sample(s) received	29 May 2023
Contact Person	Nicolette Forbes Managing Director & Senior Scientist	Tel. No.	031 572 2705
Email	nicolette@mer.co.za		

Background

As indicated earlier we do not have reference standards available for all the pesticides but were able to screen for presence/ absence using data published in the open literature. This is done based upon the technique used (LC-MS/MS) and the specific instrument model utilised whereby the unique molecular transitions creating daughter ions from the parent ion (under specific conditions), can be predicted, and hence monitored. For confirmation it would be preferable to run an authentic standard and verify both the retention time and the molecular fragmentation under the applied MS/MS conditions.

Accurate quantification without an authentic standard is unfortunately not possible. In some of our initial reports, for some pesticides such as Tebuthiuron, a structurally similar compound such as Acetamiprid was used as reference. However, the results obtained in this manner are considered tentative at best.

Description of samples received:

Purchase Order: (UPL21/21)

VM No.	Date	Project Code	Site Code	Type
31489/01	22/05/2023	UPL21/21	PCD	Water
31489/02	22/05/2023	UPL21/21	PCD03UV	Water
31489/03	22/05/2023	UPL21/21	PCDMET	Water
31489/04	22/05/2023	UPL21/21	TRIB	Water

Analysis requested: Multi residue pesticide analysis using LC-MS/MS

Method outline:

Each sample was prepared using dispersive liquid-liquid extraction.

• **Water samples**

Approximately 21 g was charged with 9 ml of acetonitrile and vortex mixed for 10 minutes whereafter phase separation was achieved through the addition of sodium chloride and magnesium sulphate. The mixture was centrifuged, and an aliquot of the acetonitrile layer was analysed.

• **Sediment samples**

Approximately 7,5 g was charged with 9 ml of acetonitrile and vortex mixed for 10 minutes. A mixture of sodium chloride and magnesium sulphate was added and further mixed. The suspension was centrifuged, and an aliquot of the acetonitrile layer was analysed.

V&M Analytical Toxicology Laboratory Services (Pty) Ltd

83 Victoria Street, George, 6529 • PostNet Suite 254, P. Bag X6590, George, 6530
Tel: +27 (0) 44 874 8484/ 873 4153 • Fax: +27 (0) 86 725 8919
Mobile: +27 (0) 82 520 2210/ +27 (0) 82 775 6932
Accurate • Reproducible • Cost effective • **RESULTS** • On time-Every time



Key to analysis results:

	Reference standard available
	Reference standard was not available, so can only report as Present (P) based on MRM from literature
0	A reading of zero indicates that the analyte was not detected (ND) No response (peak) detected under applied method conditions
<30	A reading of <30 ng/g should be regarded as positive (present), but at less than the method quantification limit (MQL). The MQL for each of the compounds listed is at least 30 ng/g or lower.
AC	Above Calibration. Sample to be re-run, results to be updated.

Units of measure (UOM): nanograms per gram (ng/g)

Analysis results:

Date Tested: 2023/06/02

Test Method: WIN-VM-061

V&M Analytical Toxicology Laboratory Services (Pty) Ltd

83 Victoria Street, George, 6529 • PostNet Suite 254, P. Bag X6590, George, 6530
 Tel: +27 (0) 44 874 8484/ 873 4153 • Fax: +27 (0) 86 725 8919
 Mobile: +27 (0) 82 520 2210/ +27 (0) 82 775 6932
 Accurate • Reproducible • Cost effective • **RESULTS** • On time-Every time

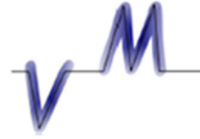


Active	31489/01	31489/02	31489/03	31489/04
2,4,5-T	0	0	0	0
2,4-D	0	0	0	0
Acetamiprid	0	0	0	0
Acetochlor	0	0	0	0
Alachlor	0	0	0	0
Ametryn	0	1	0	1
Amicarbazone	240	20	0	9
Atrazine	0	0	0	0
Azoxystrobin	1	0	0	0
Bromoxynil	0	0	0	0
Carbendazim	0	0	0	0
Carbofuran	0	0	0	0
Chlorothalonil	0	0	0	0
Chlorpyrifos	0	0	0	0
Chlorpyrifos methyl	0	0	0	0
Clomazone	0	0	0	0
Clothianidin	0	0	0	0
Cyhalothrin	0	0	0	0
Cypermethrin	0	0	0	0
Deltamethrin	0	0	0	0
Dichlorvos	0	0	0	0
diquat	0	0	0	0
Diuron	0	0	0	0
EPTC	3	0	0	0
Halosulfuron methyl	0	0	0	0
Hexaconazole	0	0	0	0
Imidacloprid	0	0	0	0
Indoxacarb	0	0	0	0
Iprodione	0	0	0	0
MCPA	0	0	0	0
Metalaxyl	0	0	0	0
Methamidophos	0	0	0	0
Methomyl	0	0	0	0
Metolachlor	216	19	0	7
Oxamyl	0	0	0	0
paraquat	0	0	0	0
Pendimethalin	0	0	0	0
Picloram	0	0	0	0
Propiconazole	0	0	0	0
Pyrimethanil	0	0	0	0
TCPY	0	0	0	0
Tebuconazole	2	0	0	0
Tebuthiuron	234	28	4	19
Terbufos	0	0	0	0
Terbutylazine	0	0	0	0
Thiamethoxam	0	0	0	0
Triclopyr	0	0	0	0

A reading of zero indicates that the analyte was not detected (ND). A reading of <30 ng/g should be regarded as positive (present), but at less than the method quantification limit (MQL).

V&M Analytical Toxicology Laboratory Services (Pty) Ltd

83 Victoria Street, George, 6529 • PostNet Suite 254, P. Bag X6590, George, 6530
Tel: +27 (0) 44 874 8484/ 873 4153 • Fax: +27 (0) 86 725 8919
Mobile: +27 (0) 82 520 2210/ +27 (0) 82 775 6932
Accurate • Reproducible • Cost effective • **RESULTS** • On time-Every time



Should you have any queries about the test results, please contact V&M-ATLS.

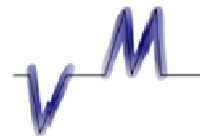
Kind regards

Mauritz Wentzel (Ph. D.) Pr. Sci. Nat.

This report relates to the specific sample(s) as received and tested as identified herein. It does not imply V&M ATLS approval of the quality of the samples in question and the test results do not apply to any similar sample(s) that has (have) not been tested.
This report may not be reproduced, except in full, with the written approval of V&M ATLS. A copy of the Confidentiality Policy is available on request.

V&M Analytical Toxicology Laboratory Services (Pty) Ltd

83 Victoria Street, George, 6529 • PostNet Suite 254, P. Bag X6590, George, 6530
Tel: +27 (0) 44 874 8484/ 873 4153 • Fax: +27 (0) 86 725 8919
Mobile: +27 (0) 82 520 2210/ +27 (0) 82 775 6932
Accurate • Reproducible • Cost effective • **RESULTS** • On time-Every time



CONFIDENTIALITY CAUTION

This message is intended only for the use of the individual or entity to which it is addressed and contains information that is privileged and confidential. If you have received this communication in error, please notify the sender of the e-mail immediately by return e-mail and delete it and any attachment(s) from your system immediately. If you are not the intended recipient, you are also hereby notified that any dissemination, distribution or copying of this communication is strictly prohibited.

TEST REPORT			
Date	01 June 2023	Report No.	230601 VM31489e
To	Marine & Estuarine Research (MER)	Sample(s) received	29 May 2023
Contact Person	Nicolette Forbes Managing Director & Senior Scientist	Tel. No.	031 572 2705
Email	nicolette@mer.co.za		

Description of samples received:

Purchase Order: (UPL21/21)

VM No.	Date	Project Code	Site Code	Type
31489/01	22/05/2023	UPL21/21	PCD	Water
31489/02	22/05/2023	UPL21/21	PCD03UV	Water
31489/03	22/05/2023	UPL21/21	PCDMET	Water
31489/04	22/05/2023	UPL21/21	TRIB	Water

Analysis requested: Quantitative determination of selected elements by ICP-MS

Water samples

Analysis results:

Date Tested: 2023-06-01

Test Method: WIN-VM-085

VM No.	Date	Project Code	Site Code	Mn (ng/ml)	Cu (ng/ml)	Zn (ng/ml)	As (ng/ml)
31489/01	22/05/2023	UPL21/21	PCD	28,3	5,9	129,0	6,9
31489/02	22/05/2023	UPL21/21	PCD03UV	17,3	2,9	171,7	4,3
31489/03	22/05/2023	UPL21/21	PCDMET	1,3	2,8	142,6	4,0
31489/04	22/05/2023	UPL21/21	TRIB	774,7	3,7	98,0	1,2

LOD – Limit of detection, LOQ – Limit of quantification

Note:

- A reading of between LOQ and LOD is reported as <LOQ

Limit of quantification (ng/ml)

Matrix	Mn	Cu	Zn	As
Water	1,0 ng/ml	1,0 ng/ml	1,0 ng/ml	1,0 ng/ml

Should you have any queries about the test results, please contact V&M-ATLS.

Kind regards

Mauritz Wentzel (Ph. D.) Pr. Sci. Nat.

*This report relates to the specific sample(s) as received and tested as identified herein. It does not imply V&M ATLS approval of the quality of the samples in question and the test results do not apply to any similar sample(s) that has (have) not been tested.
This report may not be reproduced, except in full, with the written approval of V&M ATLS. A copy of the Confidentiality Policy is available on request.*