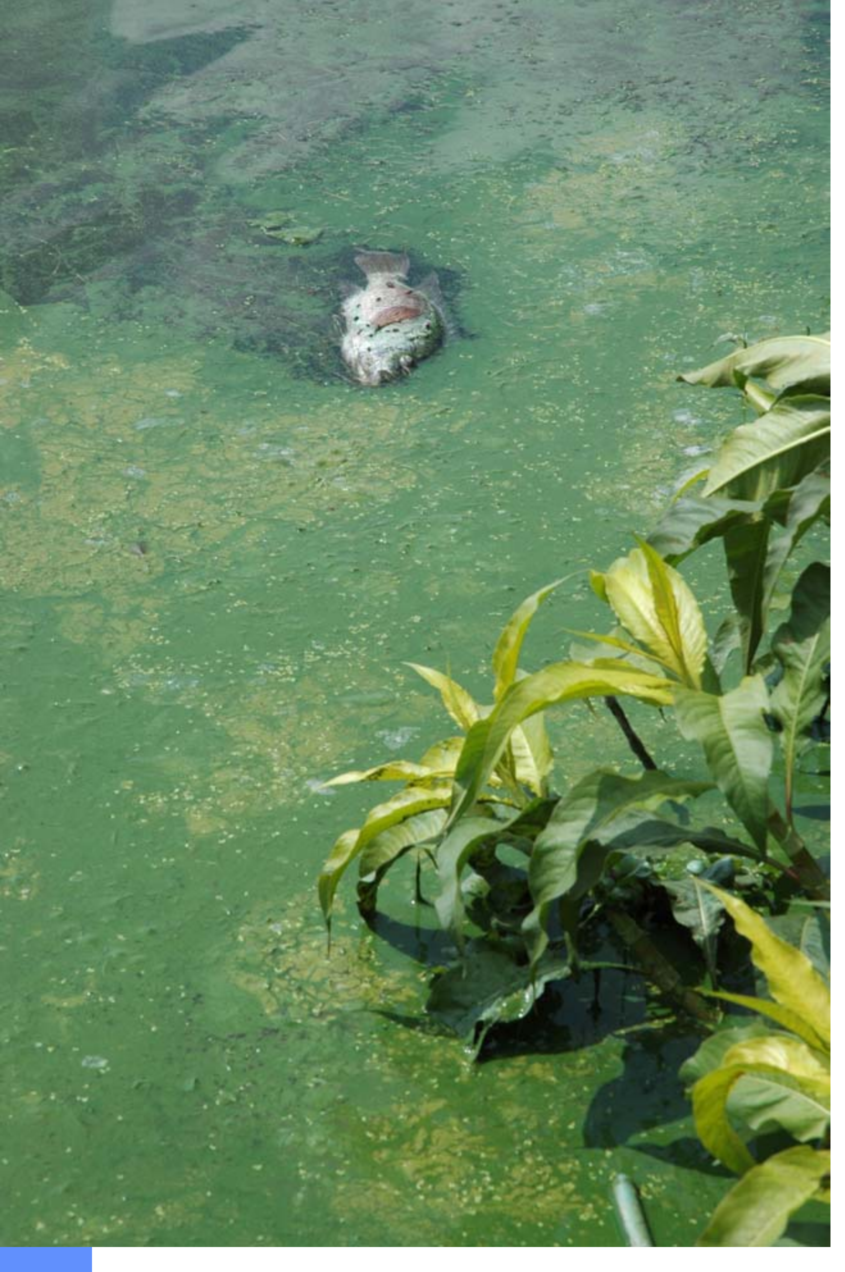
FISH KILL IN THE ROODEPLAAT DAM FROM 11th OCTOBER 2004



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DOCUMENT STATUS AND APPROVAL

TITLE: *Fish Kill in the Roodeplaat Dam from 11^{th} October 2004*

REPORT NUMBER: *N/0000/00/DEQ/0804*

AUTHORS: *B.C. Hohls and C. van Ginkel*

STATUS OF REPORT: *Final*

DATE: *December 2004*

Recommended

Mr Bonani R. Madikizela
Deputy Director: RQS

Approved

Mr Mbangiseni P. Nepfumbada
Manager: WRIM

This report must be quoted as:

Hohls, B.C., and van Ginkel, C. (2004). Fish Kill in the Roodeplaat Dam from 11^{th} October 2004. Report No. N/0000/00/DEQ/0804. Resource Quality Services, Department of Water Affairs and Forestry, Pretoria, South Africa.

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1. Introduction and Background

The first report of a fish kill was received from Mr A. Seloana from RQS on Monday the 11th October 2004. A request was received by Ms C. van Ginkel of Resource Quality Services from Ms A. Gouws of the Roodeplaat Nature Reserve late on the 14th October 2004 to investigate the reported dead fish - *Oreochromis mossambicus* (tilapia/blou kurper) - in the Roodeplaat Dam. It is uncertain when the fish kill actually commenced.

Mr J. Daffue, of the DWAF Gauteng Regional Office, was informed of the fish kill and notified of the RQS's intention to assist with the water quality sampling component of the fish kill investigation. Mr B. Hohls and Ms C. van Ginkel conducted the investigation on 15th October 2004, and undertook a follow-up sampling event on 20th October 2004 once the results of the initial water quality analyses were available.

It would appear that only adult *O. mossambicus* were affected, however, since various clean-up efforts had already been undertaken, it can not be said with complete certainty whether there were any other species involved or not. Two locations on the banks of the Roodeplaat Dam were visited during the investigation:

Site 1. **Barbergat,** at Latitude: 25° 37' 30.2" South; and Longitude: 28° 20' 40.9" East, and
Site 2. **Pienaars Inflow,** at Latitude: 25° 38' 48.5" South; and Longitude: 28° 20' 28.3" East.

On the 15th October 2004 numerous dead *O. mossambicus* were evident, however, none of them were newly dead or dying. For this reason, no fish samples were taken to the Onderstepoort laboratories for pathological examination on this date since fish putrefy soon after dying and are, therefore, then not suitable for examination once dead.

At the time of the investigation, anglers were busy catching barbel (*Clarius gariepinus*) that appeared to be unaffected by the fish kill. Live *Daphnia* were also visible in the sample bottles that were being collected for toxicity testing.

A follow-up investigation was conducted by Mr B. Hohls and Ms C. van Ginkel on the 20th October 2004, on which occasion training was given to Ms. P. Mphumbude (also of RQS) on the procedure for conducting water quality sampling for fish kill investigations. The follow-up investigation was decided upon after receiving analytical results that showed alarmingly high concentrations of certain water quality attributes, notably, unionised ammonia (NH_3-N) and dissolved zinc (Zn).

While conducting the follow-up investigation, a dying adult *O. mossambicus* of approximately 400 mm total body length was visible near Site 1. It was swimming lethargically on its side at the surface of the water. It was captured and delivered to VetPath (commercial veterinary pathologists situated at Onderstepoort) still in a live state for the purpose of conducting pathological examination.

Water quality samples and physical measurements described in the following sections of the report were taken at the locations indicated in Figure 1.

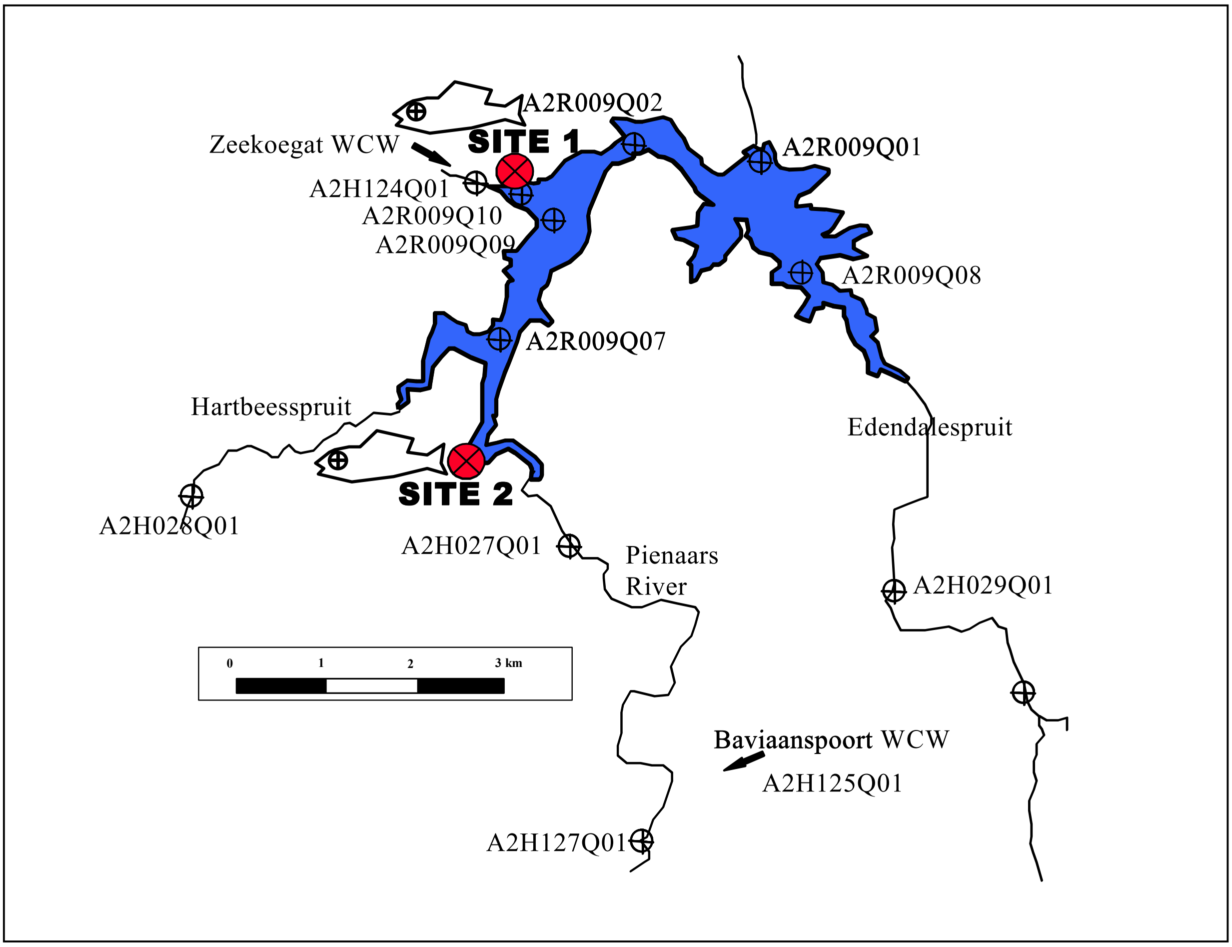

Figure 1. Diagrammatical representation of the Roodeplaat Dam indicating routine sampling sites and the fish kill investigation sites (1 and 2)

Mr J. Daffue, the Gauteng North DWAF Regional Water Pollution Control Officer, indicated during a telephone conversation with Mr. B. Hohls that there had been problems with the discharge from the Baviaanspoort Water Care Works (WCW) for the week or so prior to the fish kill taking place.

Observations made during field visits also highlighted the presence of cyanobacterial (*Microcystis*) scums in the vicinity of Site 2, especially. These scums had the characteristic blue and white colouration that is usually found during toxic events of *Microcystis*.

The following sections outline the sampling and various analyses that were conducted at the two sites. This is followed by conclusions and recommendations made by the authors.

2. Sampling and Analysis Conducted

Various water quality samples were taken at the two sites indicated in Figure 1. These samples were used to determine the concentrations of major inorganic constituents, micro inorganic constituents including trace metals, and the toxicity of water samples. Water temperature, pH, electrical conductivity (EC), and dissolved oxygen readings were taken on site. An adult *O. mossambicus* that was seen to be in difficulty on the 20th October 2004 was captured and was taken to the commercial pathology laboratory (VetPath) at Onderstepoort for pathological examination.

3. Analytical Results and Discussion

Detailed results of the various water quality samples and measurements taken at the three sites are listed in the following sections.

3.1 Physical Measurements

Physical measurements were taken of as many attributes as possible with the available field instrumentation, specifically: temperature; dissolved oxygen; EC and pH. These attributes are prone to change prior to analysis in the laboratory and, therefore, need to be assessed *in situ*. The values for these attributes are listed in Table 1.

Table 1. Measurements taken at the three sites on 15th and 20th October 2004

Attribute	Site 1 Barbergat on 15th October 2004	Site 1 Barbergat on 20th October 2004	Site 2 Pienaars Inflow on 20th October 2004
Temperature (°C)	21.97	25.6	24.1
Dissolved Oxygen (%)	77.2	109.7	123.0
Dissolved Oxygen ($mg.\ell^{-1}$)	6.74	7.90	9.13
pH	7.75	7.95	9.12
EC ($mS.m^{-1}$)	65	62	50.5
Latitude of sampling site	25° 37’ 30.2” S	25° 37’ 30.2” S	25° 38’ 48.5” S
Longitude of sampling site	28° 20’ 40.9” E	28° 20’ 40.9” E	28° 20’ 28.3” E

Dissolved oxygen concentrations of 80 – 120 % of saturation are considered to constitute the Target Water Quality Range for aquatic ecosystems (DWAF, 1996). The minimal allowable dissolved oxygen values according to DWAF (1996) are not less than 60 % for sub-lethal effects and not less than 40 % for lethal effects, respectively. The sub-lethal value relates to the 7-day mean minimum, and the lethal value relates to the 1-day minimum. According to DWAF (1996), both the 7-day minimum and the 1-day minimum should be used together. It is stated that the violation of these minimum values is likely to cause acute toxic effects on aquatic biota.

Assuming that the temperature and dissolved oxygen readings taken during the investigation are representative of those during the preceding days and weeks, it is not expected that the fish would have been under stress as a result of oxygen levels.

It is notable that the pH level at Site 2 is significantly higher than that at Site 1 (there is less difference in the laboratory-measured values presented in Table 2).

3.2 Major inorganic constituents

The major inorganic constituents analysed by the RQS laboratories are listed in Table 2, together with aquatic ecosystem guidelines for selected attributes (DWAF, 1996).

Table 2. Major inorganic constituent results of samples taken

Constituent	Site 1. 15/10/2004	Site 1. 20/10/2004	Site 2. 20/10/2004	Aquatic Ecosystem Guideline/ Category
pH	7.6	8.5	8.7	pH change should not be > 5 %
Kjeldahl nitrogen as N	10.06	6.611	3.818	Hypertrophic at Site 1 on 15/10, but Eutrophic at both Sites on 20/10
Ammonium (NH_4^+) as N	7.07	2.825	2.181	Refer to Table 3 for the conversion to UIA
Nitrate + nitrite as N	5.66	5.983	2.074	
Fluoride as F	0.3	0.266	0.290	TWQR $\leq$ 0.75
Alkalinity as $CaCO_3$	112	104	121	
Sodium as Na	66	58.5	45.5	
Magnesium as Mg	8	10.9	14.7	
Silicon as Si	6.8	6.3	4.3	
Total phosphate as P	0.752	0.433	0.336	Hypertrophic at Sites 1 & 2 on both dates
Orthophosphate as P	0.428	0.177	0.242	Hypertrophic at Site 1 on 15/10 and Eutrophic at Sites 1 & 2 on 20/10
Sulphate as SO_4	66	52.7	41.8	
Chloride as Cl	69	68.2	58.6	
Potassium as K	12.4	10.9	8.8	
Calcium as Ca	34	29.8	26.1	
EC ($mS.m^{-1}$ at 25 °C)	67.2	58.7	52.3	
Hardness	118			

Concentrations are in $mg.\ell^{-1}$ except for pH and EC

The un-ionised ammonia (UIA or NH_3-N) concentration was calculated according to the method of Wade (1999) using the NH_4^+ concentration, pH, temperature, and electrical conductivity (EC) values. On the 15th October 2004 the UIA (NH_3) concentration at Site 1 was 0.142 $mg.\ell^{-1}$ NH_3 as N (refer to Table 3). The Target Water Quality Range for UIA is less than 0.007 $mg.\ell^{-1}$ NH_3-N, with the chronic effect value being 0.015 $mg.\ell^{-1}$, and the acute effect value being 0.100 $mg.\ell^{-1}$ NH_3-N. The concentration was, therefore, above the acute effect value for an aquatic ecosystem (DWAF, 1996). Therefore, it is highly likely that unionised ammonia was implicated in the fish kill.

Table 3. **Calculation of UIA according to the method of Wade (1999)**

Site and date of sampling	pH	NH_4 (mg/l)	Temp (C)	EC (mS/m)	UIA (NH_3 mg/l)
Site 1 "Barbergat" 15/10/2004	7.6	7.07	21.97	67.2	**0.142**
Site 1 "Barbergat" 20/10/2004	7.95	2.825	25.6	62	**0.154**
Site 2 "Pienaars Inflow" 15/10/2004	9.12	2.181	24.1	50.5	**0.952**

On the 20th October, however, it can be seen that in spite of the NH_4 concentration decreasing, UIA (NH_3) concentrations were still beyond the AEV, especially at Site 2 at the Pienaars Inflow. This is largely due to the increased pH, especially at Site 2, and higher water temperatures resulting in higher percentages of the available NH_4 (as N) being in the toxic NH_3 (UIA) form. All of the UIA concentrations are above the AEV, and at Site 2 on the 20th October 2004 it was very far above the AEV.

Other attributes at high concentrations were: KN as N; NO_3+NO_2 as N; TP as P; and PO_4 as P. These indicate a highly nutrient enriched system that may have been exacerbated by the problems associated with the discharge from the Baviaanspoort WCW. Gauging from samples taken on the 20th October, it would appear that the source of contamination is from higher up in the Pienaars River rather than within the dam itself.

3.3 Acute Toxicity Tests

A sample was taken for toxicity analysis at Site 1 on the 15th October 2004. The toxicity tests using 4-11 day old *Poecilia reticulata* fish (guppies) indicated no toxicity present in the water. This is surprising, but consistent with the observation of juvenile fish alive and apparently unaffected in the shallow waters. However, the algal growth inhibition test with *Selenastrum capricornutum* showed an inhibitory effect of 82 %. This puts the sample into the Class III Acute Hazard level. The algal inhibition test, therefore, indicates lethal conditions within the Roodeplaat Dam at the time of the fish kill. It is remarkable though that only dead adult fish were evident.

It is, however, unclear why there is a discrepancy between the results of the fish toxicity and algal inhibition tests.

3.4 Trace Metal Analyses

Samples for trace metal analysis were taken at both sites (only at Site 1 during the initial investigation but both sites at the follow-up investigation) to determine whether trace metal concentrations could have been at levels that would have resulted in, or contributed to, the fish kill. The results of the analyses appear in Table 4.

Table 4. Trace metal concentrations recorded at the two sites

Constituent (mg.ℓ$^{-1}$)	Limit of quantitation	Site 1. 15/10/04	Site 1. 20/10/04	Site 2. 20/10/04
B – dissolved	0.026	< 0.026	0.103	0.066
Al – dissolved	0.182	< 0.182	< 0.182	< 0.182
V – dissolved	0.022	< 0.022	< 0.022	< 0.022
Cr – dissolved	0.016	< 0.016	< 0.016	< 0.016
Mn – dissolved	0.012	0.123	0.036	< 0.012
Fe – dissolved	0.027	< 0.027	< 0.027	< 0.027
Ni – dissolved	0.032	< 0.032	< 0.032	< 0.032
Cu – dissolved	0.044	< 0.044	< 0.044	< 0.044
Zn – dissolved	0.014	**0.222**	< 0.014	< 0.014
Sr – dissolved	0.004	0.082	0.086	0.099
Mo – dissolved	0.037	< 0.037	< 0.037	< 0.037
Cd – dissolved	0.009	< 0.009	< 0.009	< 0.009
Ba – dissolved	0.014	< 0.014	< 0.014	< 0.014
Pb – dissolved	0.126	< 0.126	< 0.126	< 0.126

Mr P. Botes (Botes, 2004) indicated that the zinc (Zn) concentration on the 15th October 2004 was higher than he had ever seen for the Roodeplaat Dam in previous analyses, and it is above the Acute Effect Value (AEV) of 0.036 mg.ℓ$^{-1}$ Zn (DWAF, 1996) by a significant margin. It is, therefore, also likely that the elevated Zn levels played a role in the fish kill and may be indicative of the nature of the contaminant source. Zinc is usually associated with industrial activity. It is used extensively in the following industries: metal galvanising; dye manufacture and processing; pigments (paints and cosmetics); pharmaceuticals; and fertiliser and insecticide (DWAF, 1996).

The Zn concentration had once again fallen to a concentration less than the level of quantitation by the 20th October 2004.

3.5 *Post Mortem* Examination

The following are the ***post mortem* findings** of Du Plessis (2004) relating to the adult *Oreochromis mossambicus* delivered to VetPath for pathological examination:

1. Female gravid individual in moderate body condition, mild to minimal autolysis (death of cells) present.
2. The liver is orange to yellow-brown in colour, mildly enlarged and reveals multifocal (± 4) yellow nodules therein, varying from 5-8 mm in cross section (nodules can be indicative of a situation where the liver regenerates in an abnormal pattern after chronic disease).
3. Moderate to severe renal congestion. This is apparently indicative of heavy metal toxicity (Kempster, 2004).
4. Moderate splenic congestion with mild splenomegaly (enlargement of the spleen as a result of bacterial/parasitic/viral infections).
5. The stomach is empty and the pylorus area (stomach outlet) is slightly hyperplastic (excessive number of normal cells).
6. The intestinal tract contains watery bile-stained fluid, and in the caudal intestinal tract the intestines have very thin walls, which are transparent, revealing the

abundant watery orange to red contents in the lumen (the inner part of the intestines – the space).

7. The gills are mildly congested.
8. Wet-prep smears from the gills and skin did not reveal notable parasites although these disappear soon after death of the fish and may thus be artificially absent

Histological findings:

Small intestine: Autolysis (cell death) is already advanced and makes a thorough histological investigation of the intestinal tract impossible. In the surrounding fat tissue perivascular aggregates of pancreatic tissue are visible, and appear normal. Further sections from the intestinal tract revealed more well-preserved intestinal loops in which specific histological lesions were absent.

Spleen: Moderate lymphoid hyperplasia (excessive growth of normal cells resembling lymph tissue that can exert pressure on other organs/tissues) with multifocal aggregates of haemosiderin (a granular brown substance composed of ferric oxide left over from the break down of haemoglobin) pigment-containing macrophages (certain types of white blood cells) especially around blood vessels.

Hepatopancreas (primitive form of digestive gland that contains cells similar to those found in mammalian liver): The macroscopically visible yellow nodules, histologically revealed necrogranulomas (nodules of inflammatory cells) with central necrotic caseous (damaged or necrotic tissue) centres surrounded by mononuclear leukocytes (a type of white blood cell). Causative organisms were not visible within these granulomas.

Gill: Multifocal secondary lamellae appear hypercellular with mildly increased numbers of mononuclear leukocytes (a type of white blood cell - mostly lymphocytes, a type of white blood cell that attacks bacteria and viruses). This confirms a mild proliferative branchiitis. No pathogenic organisms including protozoa were visible in association with the gill lesions.

Kidney: Multifocal small aggregates of haemosiderin pigment containing macrophages are visible in the interstitium (supporting tissue). Scattered renal tubular epithelial cells contain eosinophilic protein globules within the cytoplasm. The cells are however intact and furthermore appeared normal.

The other organs examined did not reveal any notable histological lesions including the ovary, stomach, eye, omental fat (deep abdominal fat) and heart muscle.

***Post mortem* discussion**

The necrogranulomas within the liver were few and small, and probably represent an incidental finding of an underlying chronic infectious condition within the hepatic parenchyma. The other organs did not reveal signs of underlying infection, mild inflammation throughout the intestinal tract is normally seen due to antigenic stimulation from flora within the intestines.

Mild profilerative branchiitis is not a specific lesion coupled with a specific condition, but it is mostly associated with poor water quality, which may include high ammonia levels, high nitrites and nitrates as well as decreased dissolved oxygen concentration. Low dissolved oxygen concentrations may develop due to cloudy, overcast skies and rain, as well as algal die-off. Organic contaminants within the dam may lead to increased ammonia, nitrates and nitrites as a decrease in oxygen tension.

3.6 Other Possible Factors

Mr J. Daffue (Daffue, 2004) reported that there had been process problems at the Baviaanspoort WCW for a number of weeks leading up to the fish kill. This may go some way towards explaining the ongoing high UIA concentrations recorded.

Cyanobacterial (*Microcystis*) scums were evident, especially at Site 2. These could have given off toxins that also affected the fish. The total microcystin concentration that was measured at the Babergat site (Site 1) on the 15th October 2004, however, was 0.3 µg.ℓ^{-1}. This concentration is well within acceptable limits and should not have contributed to the fish kill.

Routine major inorganic constituent sampling within the Roodeplaat Dam revealed that when the fish kill was initially reported by Mr A. Seloana (of RQS), the levels of certain chemical attributes were already elevated to the point where they would have been problematical. Refer to Table 5 for attribute concentrations as downloaded from the Water Management System (WMS) for selected monitoring sites with the Roodeplaat Dam. For the location of the various sample sites listed in Table 5, please refer to Figure 1 near the beginning of the report.

Table 5. Major inorganic constituent results of routine samples taken at the Roodeplaat Dam

Constituent	A2R009Q01 11/10/04	A2R009Q02 11/10/04	A2R009Q07 11/10/04	A2R009Q08 11/10/04	A2R009Q09 11/10/04	A2R009Q10 11/10/04
pH	9.105	8.87	8.721	8.969	8.701	No data
Kjeldahl nitrogen as N	1.166	1.848	3.203	1.199	2.566	No data
Ammonium (NH_4^+) as N	0.273	0.431	1.424	0.153	0.972	No data
Calculated NH_3 as N	**0.108**	**0.118**	**0.302**	**0.049**	**0.199**	
Nitrate + nitrite as N	1.744	1.811	1.902	1.455	2.088	No data
Fluoride as F	0.284	0.279	0.288	0.325	0.279	No data
Alkalinity as $CaCO_3$	113	115	124	112	120	No data
Sodium as Na	41.8	42.8	44.7	42.6	44.0	No data
Magnesium as Mg	15.1	15.3	15.1	15.3	14.8	No data
Silicon as Si	2.8	3.1	3.9	2.7	3.6	No data
Total phosphate as P	0.124	0.158	0.264	0.088	0.257	No data
Orthophosphate as P	0.068	0.086	0.166	0.043	0.114	No data
Sulphate as SO_4	37.2	38.7	38.7	38.5	41.6	No data
Chloride as Cl	50.5	48.3	51.4	45.4	50.0	No data
Potassium as K	8.1	8.2	8.6	8.0	8.5	No data
Calcium as Ca	25.7	26.1	29.8	24.7	28.9	No data
EC (mS.m^{-1} at 25 °C)	47.7	48.9	52.3	48.2	50.7	No data
Hardness	127	128	137	125	133	No data

Concentrations are in mg.ℓ^{-1} except for pH and EC

It can be seen that even prior to the earliest known date of the fish kill (the 11th October 2004) the levels of certain attributes were high enough to result in distress to fish, if not the likelihood of death.

Specifically, the NH_3-N concentrations at all locations, except A2R009Q08 (Eastern/ Edendalespruit arm of the dam), were above the AEV. These values were calculated based on the assumption of a similar water temperature on the 11th October 2004 to that recorded on the 15th October 2004. It should be noted, however, that even a major difference in temperature (such as freezing conditions) would not have brought the calculated NH_3-N concentration below the AEV.

Site A2R009Q07 (hereafter referred to as Q07) is located closest to Site 2 of the fish kill investigation while site A2R009Q09 (hereafter referred to as Q09) is located closest to Site 1. Site A2R009Q10 (Q10) would have been closer to Site 1 and, therefore, even more ideal, but a sample was not taken at this site on the 11th October 2004.

Referring back to Table 1 and Table 2 in conjunction with Table 5, it can be seen that the NH_3-N concentration was higher at Q09 on the 11th October than it was during the initial fish kill investigation at Site 1 on the 15th October or during the subsequent investigation on the 20th October. The NH_3-N concentration at Q07, however, was considerably lower on the 11th October than it was at Site 2 on the 20th October.

4. Conclusions and Recommendations

It is clear that some contaminant has entered the water of the Roodeplaat Dam and has resulted in a significant fish kill that has persisted over a number of weeks. Based on the limited sampling conducted, it would appear that the contaminant has come from the direction of the Pienaars River inflow side of the Dam.

Since it was reported by Daffue (2004) that there were significant process problems at the Baviaanspoort WCW, which is situated approximately 10 km upstream of the Roodeplaat Dam on the banks of the Pienaars River, it would appear that this could be a major contributor to the fish kill.

It should be noted that at the time of this fish kill there were also reports of other fish kills from a wider geographical distribution (from the Lindleyspoort Dam in the North West Province to the Blesbok River in Mpumalanga). Fish kills are not uncommon for this time of year when atmospheric and water temperatures suddenly increase.

In this way the fish kill in the Roodeplaat Dam could have been exacerbated by climatic conditions specific to the time of the year.

Dr P. Kempster (Kempster, 2004) indicated that Zn and NH_3 could combine to form $Zn(NH_3)_2$ that is lipid soluble and very toxic (Dr S. Jooste (Jooste, 2004), however, says that it is very unlikely for this compound to occur in water). In Dr Kempster's opinion the presence of this complex would have been sufficient to result in a fish kill even without any other factors being involved. He further stated that the presence of living juvenile fish confirms this since the kidney and liver cells of juvenile fish are different to those of adults and there can, therefore, be a different response. The renal congestion evident in the adult fish is consistent with the damage that would have resulted when the fish's kidneys attempted to excrete the toxins.

Algal scums were very abundant and it is also possible that toxins produced or the die-off of the algae itself may have played an additional role in the fish kill.

It is recommended that an intensive investigation be conducted upon the water treatment process at the Baviaanspoort WCW to ascertain what the problem is and find means of ensuring that it doesn't recur. It is also necessary to look to the industrial activities higher up in the drainage region to determine the source of the elevated Zn levels.

5. Acknowledgements

The authors would like to acknowledge the following individuals and groups for their contributions to this report:

- The various RQS laboratory staff for the dedicated and speedy way in which they conducted the required analyses needed for this report.
- Dr P.L. Kempster and Dr S.H.J. Jooste for their insights.
- The contribution made by the VetPath at Onderstepoort.

6. References

Botes, P. (2004). Personal communication. Resource Quality Services, Department of Water Affairs and Forestry. Pretoria, South Africa.

Daffue, J. (2004). Personal communication. Gauteng Regional Office, Department of Water Affairs and Forestry. Pretoria, South Africa.

Du Plessis, L. (2004). Report on *Oreochromis mossambicus* received for *post mortem* on 20/10/04: VetPath Ref 2004-9614. VetPath Veterinary Pathologists. Onderstepoort, Pretoria.

DWAF (1996). SA Water Quality Guidelines, Aquatic Ecosystems, Department of Water Affairs and Forestry, Volume 7, 1996.

Jooste, S.H.J. (2004). Personal communication. Principal Specialist Scientist. Resource Quality Services, Department of Water Affairs and Forestry. Pretoria, South Africa.

Kempster, P.L. (2004). Personal communication. Chief Specialist Scientist. Resource Quality Services, Department of Water Affairs and Forestry. Pretoria, South Africa.

Wade, P. (1999). Calculation of un-ionised ammonia in river water samples. Council for Scientific and Industrial Research, Pretoria, South Africa. Exact reference not known.