

RESUMÉ

Of the problems currently being experienced with natural and man-made water bodies, eutrophication is one of the most important. Eutrophication is the enhancement of the natural process of biological production in rivers, lakes and reservoirs, caused by an increase in nutrient levels, usually phosphorus and nitrogen compounds. These increased nutrient levels usually result in an increased phytoplankton biomass, which is often dominated by toxic cyanobacterial species. Eutrophication has a severe impact on the water quality and impairs the use of water for drinking, industry, agriculture and recreation.

The management of a eutrophic water body usually involves treating toxic algal blooms, as well as controlling nutrient input. However, reducing nutrient input as well as the internal source is the only feasible means of long term eutrophication management, as in many shallow lakes the phosphorus accumulated in the sediment may be many times greater than that in solution. In this study, Phoslock[®], a lanthanum-modified bentonite clay capable of removing phosphorus by adsorption, was characterised in the laboratory in terms of its kinetics and the effect of initial pH and phosphorus concentration on the adsorption capacity. The product was also tested in cyanobacteria-containing lake water with a high pH value under laboratory conditions in order to gain understanding of the behaviour of Phoslock[®] in a natural water body. Phoslock[®] was most effective between pH 5 and pH 8, with a decrease in the adsorption capacity above pH 9. Furthermore, phosphorus was not released under anoxic conditions. Phoslock[®] was then tested in a field trial at Hartbeespoort Dam, and the soluble phosphorus concentration was successfully reduced from 0.2mg.l^{-1} to below 0.05mg.l^{-1} , the threshold for cyanobacterial bloom formation. Cyanobacterial growth was visible from much earlier in summer in the control area, and the bloom was more severe throughout the summer months. The low phosphorus concentration in the water body and the reduced concentration in the sediment therefore effectively reduced the incidence and severity of the algal bloom in the treated site.

Limiting the amount of phosphorus in a water body, and thus increasing the N:P ratio, was likely to affect the entire microbial community composition, not only that of the

cyanobacteria and algae. Samples were taken monthly from the Phoslock[®] field trial site between July and February, and the effect of reduced phosphorus concentration on the cyanobacterial and eubacterial community composition was examined using denaturing gradient gel electrophoresis (DGGE). Unicellular cyanobacteria were present in both the treated and control areas, but there was a lag in the appearance of these species in the treated area. The different trophic levels of the treated and control areas affected the filamentous cyanobacterial population, as filamentous species were more prevalent in the treated area during the summer months than in the control area, and the treated area had a higher species diversity. As the cyanobacteria became more dominant in the treated and control areas from October, there appeared to be a shift in the bacterioplankton population. Species of Actinobacteria and Bacteroidetes were present in both the treated and control areas only until October, with one species of Actinobacteria only being present in the treated area. From November, the bacterioplankton population was dominated by β - and δ -proteobacteria. The Phoslock[®] treatment itself did not appear to affect the bacterial population, as the treated and control areas displayed similar patterns. For both the cyanobacteria and the bacterioplankton, the greatest effect on the species composition was in fact the seasonal change from winter to summer, as expected.

A bacterial species that was isolated from Hartbeespoort Dam that appeared to have cyanobacteriolytic activity was identified as *Bacillus cereus*. The cyanobacteriolytic nature of this species against *Microcystis aeruginosa* has previously been documented in the literature. The bacteria used in this study required contact for lysis, as in previous studies, but aggregation of the cyanobacteria was reduced in treated flasks. This may indicate that the strains were different, with the lytic substance and mechanism of lysis differing between these two organisms. The critical predator-prey ratio was 1:1 (cyanobacteria to predatory bacteria), as lower ratios of bacteria to *M. aeruginosa* did not cause the cyanobacterial population to decrease, although ratios of 1:10 and 1:100 kept the cyanobacterial population steady. A 1:1 ratio reduced the cyanobacterial population by 50% over a 14 day period, even though the bacterial population was seen to double in this time. *Bacillus cereus* was able to use *Microcystis aeruginosa* as its only nutrient source. This is of great importance in terms of the formation of a biological control product, as no additional nutrients will need to be supplied to the bacteria.

The combination of this potential biological control agent with Phoslock[®] was investigated in order to determine whether the two agents could be used together to treat both the cause and symptoms of eutrophication simultaneously. When Phoslock[®] and the cyanobacteriolytic bacteria were combined in a bacterial culture, Phoslock[®] had no effect on the growth rate of the bacteria. However, when the two agents were combined to assess the possibility of synergism, treatment with both Phoslock[®] and bacteria was no more effective than bacteria alone, and Phoslock[®] alone was more effective than either treatment with bacteria or with a combination of Phoslock[®] and bacteria. There is therefore no synergistic effect when these agents are used in combination, and Phoslock[®] was the most effective treatment method.

Various flocculants have been investigated for cyanobacterial removal in wastewater treatment as well as in natural water bodies. These include synthetic organic polyelectrolytes, chitosan, and various clays. In this study, fly ash, a waste product in the burning of coal for electricity generation, was investigated as a potential cyanobacterial flocculant. Samples from seven different power stations were tested, and it was found that the ash with the smallest particle size had the highest flocculation efficiency; between 65 and 95% depending on the thickness of the algal layer. Four out of the seven fly ash samples tested caused cyanobacterial cell death after 36h. This was possibly related to the leaching of toxic elements, although only a small percentage of the total amount of trace elements were leached into solution, even at pH 2. The addition of fly ash to natural water bodies may not be hazardous, especially considering the added benefits of potential toxin removal from the water. As with the cyanobacteriolytic bacteria, field trials are necessary with the fly ash in order to determine the effect on a large body of water as well as whether the flocculation would be permanent in the turbulent conditions of a natural water body.

The various methods for remediating both the causes and symptoms of eutrophication that were investigated in this study can all potentially reduce the impact of eutrophication on natural water bodies. However, it is unlikely that any single technique used in isolation would allow a eutrophic water body to return to its natural mesotrophic state. Instead, the combination of techniques addressing both the cause and the result of eutrophication will increase the likelihood of successful remediation.

Appendix A

1. Sequences obtained from DGGE bands in Chapter 5

1.1 Partial 16S rDNA sequences obtained from from bands in the cyanobacterial specific DGGE gels, and their accession numbers in GenBank

1a (EU94509)

CAGCCAACCGCTTCGCAATGGGGTTCTTTTAAAGCCACAATTTACGCTCCC
TGGNAATTCCCTTTACTTTCTATACTCTAGTCTAATAGTTTCGACTGCGATTT
TGAAGTTGAGCTTCAAGATTTAACAGTTGACTTATTAAACCACCTACAGACG
CTTTACGCCAGTGATTCCGGATAACACTTGCATCTTCCGTCTTACCGCGGC
TGCTGGGACGGAGTTAGCCGATGCTTATTCTCCAGGTACACGTCCTTTTGTT
CCTCCCTGAAAAAAGAGGTTTACAACGCATAGGCCGGTATCCCTCAGGCGA
GATTGCTCCGTCANTTTTCAAACAATGCGGAAGTTCCCCCGGGCGAGTCGGC
CTGCCGCCGG

2a (EU94510)

GTTCGGCCCAGTACCCACGTTTCGCTATGGGGTTCTTTTCANNNATACCAAT
TTCACCGCTACACTGGGAATTCCTGCNTCTTCTACTGCTCTCTAGTCTGCCAG
TTTCCACTGCCTTTAGGTCGTTAAGCAACCTGATTTGACGGCAGACTTGGCT
GACCACCTGCGGACGCTTTACGCCAATAATTCCGGGTAACGCTTGCCTCCC
CCGTCTTACCGCGGCTGCGGGGACGGAGTTAGCCGAGGCTTATTCCTCAGGT
ACCGTCAGAACTTCTTCCCTTGAGAAAAGAGGTTTAAAATCCAAAGACCTTCC
CCCCCTCACGCGGTGTTTCCCCATCAGGTTTTCGCCATTGCGCAAAAATCC
CCCCGGGGGG

3a (EU94511)

CAGTTCGGCCCCCTACACGCTTTCGCACTGAGGATCTTNNNCNCTAGGCATTT
CACCGCTACACTGGGAATTCCTGTTACCCCTAGTGCTCTCTAGTCTGCCAGT
TTCCACTGCCTTTAGGTCGTTAAGCATCCTGATTTGACGGCAGACTTCGTTG
ACCACCTGCGGACGCTTTACGCCAATAATTCCGGATAACGCTTGCCTCCCC
CGTATTACCGCGGCTGCTGGCACGGATTTAGCCGAGGCTTATTCCTCAGGTA

CCGTCAGAACTTCTCCTTTGAGAAAAAAGGTTACAATCCAAAGCTCTTCCTC
CCTCACGCGGTGGTTCTCCCTCAGGTTTTCCCCATTGCG

4a (EU94512)

ATTTCCGCACTGGGGAAAGNAANCNCTACCCATTTACCGCTACACTGGGA
ATTCCGGCTACCCATACTGTTTTTTAGTCTGCAAGTTTCCACCGCCTTTAGGT
CGTTAAGCAACCTGATACTTGTCTGACCACCTGCGGACGCTTTACGCCAAT
AATTCCGGATAGCGTTTGCCTCCCCCGTATTACCGCGGCTGCTGGAACGAAT
TTAGACAAGGCTGATTCCTCAAGTACCGTCANAACCTTCTTCCTTGAGAAAAG
AGGGGACAATCCAACTCCTTCCTACCGACGAAATGTTTCTCGAACAGGAA
TAACCCCATTTGCGGAAAGTTCCCCCGGGCGGGGGCGG

5a (EU94513)

TTTCGCATGAGTTCTNNAACCNACGAATTTACCCTCCTGGGAATTCCTGCTA
CCCTTACTGCTCTCTAGTCTGCCAGTTTCCACCGCCTTTAGGTGGTTAAGCA
ACCTGATTTGACGGCAGACTTGGCTGACCACCTGCGGACGCTTTACGCCAA
TAATTCCGGATAACGCTTGCCTCCCCCGTATTACCGCGGCTGCTGGCACGGA
GTTAGCCGAGGCTGATTCCTCAAGTACCGTCAGAACTTCTTCCTTGAGAAA
GAGGTTACAATCCAAAGACCTTCCTCCCTCACGCGGCGTTGCTCCGTCGGGT
TTCCCCCATTTGCGAAAAATTCCCCCGGGCGGGGGCTGT

6a (EU94514)

ACTGGGGTCCTAATCCCTTGTTCGCCGGGGTTTTCTTNAANCNNAGGCTTT
ACCGCTACACCTGGATTCCTCCTGNNCTATCNCTCTCTAGTCTCACAGTTTCC
ATTGCCGATCCAAGGTTGAGCCTCGGGCTTTGACAACAGACTTATCAAACA
GCCTACGTACGCTTTACGCCAATAATTCGGGATAACGCTTGCATCCTCCGT
CTTACCGCGGCTGCTGGCACGGAGTTAGCCGATGCTTATTCGTCAGGTACCG
TCATTACCTCCCCTAACAAAAAAGGTTTACAACCCACCGGCCCTCGTCCCTC
CAACGGTTTTGTCCCCCAGGGGTTTTGCCCTTNCGAAAATTCCCC

7a (EU94515)

CCCAGTTGCACCTTGGTGTTCTGANGNGNCTCCGCATTTACCGCTACACCG
GGAATTCCTGNGNCCATATCTCTCTCTAGTCTGACAGTTTCCATTGCCGATC
CAAGGTTGAGCCTCGTGCTTTGACAACAGACTTATCAAACAGCCTACGTAC

GCTTTACGCCCAATAATTCCGGAATAACGCTTGCATCCTCCGTCTTACCGCG
GCTGCTGGCACGGAGTTAGCCGATGCTTATTGTCAGGTACCGTCATTATCTT
CCTTAACAAAAAAGGGGTACAACCCACAGGCCTTCTTCCCTCACGCGGTATT
GCTCCGTCAGAGTTTCGC

8a (EU94516)

AGTTCAGTCCAGCACCCGCTTTCACCACTGGTGTCTTGTAGAGNATACGCA
TTTCACGCTACACCGGGAATTCCTCCTGGCCTATCTATCTCTAGTCTNACAG
TTTCCATTGCCGATCTAAGGTTGAGCCTCGGGCTTTGACAACAGACTTATCA
AACAGCCTACGTACGCTTTACGCCCAATAATTCCGGATAACGGTTGCTTCCT
CCGTCTTACCGCGGCTGCTGGGACGGAGTTAGCCGATGCTTATTCGTCAGGT
ACCGTCATTATCTTCTCTAAAAAAAGAGAGAACAACCGACAGGGCTTCGT
CCCTCACGCGGGATTGCTCCCTCAGGGTTTTGCCAATAGCCCAAATTCCCC
CGGGCGGGGGGGGTGTGACCTGAGCGTGGCGCCCCGGGGAAGTTTCG

9b (EU94517)

AGTGTTAGTNATAGCCCAGTAAAGTGCCTTCGCCATCGGTGTTCTTTNNANA
NCTACGCATTTACCGCTCCACTGGAAATTCCTTTACCCCTACTATACTCTA
GTCTAATAGTTTCGACTGCTGTTTTGAGGTTAAGCCTCAAGATTTAACAGTT
GACTTATTAACCACCTACAGACGCTTTACGCCAGTGATTCCGGATAACAC
TTGCATCCTCCGTCTTACCGCGGCTGCTGGCACGGAGTTAGCCGATGCTTAT
TTTTAGGTACACGTCATTTTTTTCTCCCTGAAAAAAGAGGTTTACAACCA
GGGGGGTTTTCTCCCCACGGGGTTTTCCCCC

10b (EU94518)

GTGTCAGATACAGCCCAGTAGCACGCTTTCGCCACCGATGTTCTTCNNNNCN
CTACGCATTTACCGCTACACTGGGAATTCCTGCTACCCCTACTGCTCTCTA
GTCTGCCAGTTTCCACCGCCTTTAGGTCGTTAAGCAACCTGATTTGACGGCA
GACTTGGCTGACCACCTGCGGACGCTTTACGCCCAATAATTCCGGATAACGC
TTGCCTCCCCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTGAT
TCCTCAAGTACCGTCAGAACTTCTTCTTGAGAAAAGAGGTTTACAATCCAA
AGACCTTCTCCCTCACGCGGCGTTGCTCCGTCAGGCTTTCGCACATTGCGG
AAAATTCCCC

11b (EU94519)

AGTGTACAGATACAGCCCAGTAGCACGCTTTCGCCACCGATGTTCTTCCNANN
CNCTACGCATTTACCGCTACACTGGGAATTCCTGCTACCCCTACTGCTCTC
TAGTCTGCCAGTTTCCACCGCCTTTAGGTGCTTAAGCAACCTGATTTGACAg
CAGACTTGGCTGACCACCTGCGGACGCTTTACGCCAATAATTCCGGATAAC
GCTTGcCTCCCCGTATTACcGCGGCTGctGGcACGgAGTTAGccgAgGcTgATTC
ctCAaGTACCGtCaGAaCTTCTTCCtTGAGAAAAGAGGtTTACAATCCAAAGACC
TTCcTCCCTCCcGcGGCGTTGCTCCGTACAGgcTTTCGCccATTGCGGAAAATTCC
CCCGGGcGGG

12b (EU94520)

GTCAGATACAGCTCAGTAGCAGCTTTCGCCACCGATGTTCTTCNAANCTCTA
CCATTTTACCGCTACCTGGGAATTCTGCTATCCTACTGCTCTCTAGTCTGCCA
GTTTCCACCGCCTTTAGGTGGTTAAGCCACCTGATTTGACAGCAGACTTGGC
TGACCACCTGCGGACGCTTTACGCCAATAATTCCGGATAACGCTTGCCTCC
CCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTTATTCCTCAAG
TACCGTCAGAACTTCTTCCCTTGAGAAAAGAGGTTTACAATCCAAAGACCTTC
CTCCCTCACGCGGCGTTGCTCCGTACAGGTTTTCGCCCATGCGGAA

13b (EU94521)

GTGTCAGATACAGCCCAGCAGGACGCTTTCGCCACTGGTGTCTTCCCAATA
TCTACGCATTTACCGCTACACTGGGAATTCCTGCTGCCCTACTGCTCTCTA
GTCTGCCAGTTTCCACTGCCTTTAGGAGGTTAAGCATCCTGATTTGACAGCA
GACTTGTCTGACCGCCTACGGACGCTTTACGCCAATAATTCCGGATAACGC
TTGCCTCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTGAT
TCCTCAGGTACCGTCAGAAATTTTTCTTTGAGAAAAGAGGTTTACAATCCAG
AGATCTTTCTCCCTCACGCGGTGGTGTCTCCCTGAGGTTTTCCCTAT

14b (EU94522)

GTCAGATACAGCCCAGTAGGACGCTTTCGCCACTGGTGTCTTCNGAAANCT
ACGCATTTACCGCTACACTGGGAATTCCTGCTGCCCTACTGCTCTCTAGT
CTGACAGTTTCCACTGCCTTTAGGAGGTTAAGCCTCCTGATTTGACAGCAGA
CTTATCAAACCGCCTACGGACGCTTTACGCCAATAATTCCGGATAACGCTT
GCCTCCTCCGTCTTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTTATTC

CTCAGGTACCGTCAGAATTTCTTCCTTGAGAAAAGAGGTTTACAATACAAA
GACTTTCCTCTCTCACGCGGTGGTTCTCCCTGGGGTTTTCC

15b (EU94523)

GTCAGATACAGCCCAGCAGGACGCTTTCGCCACTGGTGTTCCTCCAGAATCT
ACGCATTTACCGCTACACTGGGAATTCCTGCTNCCCCTACTGCTCTCTAGT
CTGACAGTTTCCACTGCCTTTAGGAGGTTAAGCATCCTGATTTGACAGCAGA
CTTATCAAACCACCTACGGACGCTTACGCCAATAATTCCGGATAACGCTT
GCCTCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTTATTC
CTCAGGTACCGTCAGAATTTTTTCTTTGAGAAAAGAGGTTTACAATACAAAAG
ATCTTCCCCTCTCACGCGGTGGTTCTCCCTGAGGTTTTCCC

16b (EU94524)

GTGTCAGATACAGCCCAGTAGCACGCTTTCGCCACCGATGTTCTTCCCAATC
TCTACGCATTTACCGCTACACTGGGAATTCCTGCTACCCCTACTGCTCTCTA
GTCTGCCAGTTTCCACCGCCTTTAGGTCGTTAAGCAACCTGATTTGACGGCA
GACTTGGCTGACCACCTGCGGACGCTTACGCCAATAATTCCGGATAACGC
TTGCCTCCCCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGAGGCTGAT
TCCTCAAGTACCGTCAGAACTTCTTCCTTGAGAAAAGAGGTTTACAATCCAA
AGACCTTCTCCCTCACGCGGCGTTGCTCCGTCAGGCTTTCGCCCATGCGG
AANATTCCCCCGGGCGGGG

1.2. Partial 16S rDNA sequences obtained from from bands in the general bacterial DGGE gel

1. (EU94525)

TACAGCGGCTGCTGGCCATGGTGAGCATGTATTACCGCGGCTGCTGGCCAA
TGGTGAGCATGTATTACCGCG

2 (EU94526)

ATGGCAGCGGCGGACGGGTGCGTNANNNNNNNNNNNNNNTGAGGTGGGG
GACAACCCTGGAAANGGGGCTAATACCGCATATGGGCTGAGGCCCAAAGCC
GAGAGGGGNNTTAGGAGCGGCCTGCGTCCGATTAGCTAGNNGGNGGGGAA
GGCCTACCAAGGCTCCGATCGGNAGCTGGTCTGAGAGGCGATCAGCCACAC

TGGGACTGAGACACGGCCCAGACTCCTACGGGAGGCAGCAGTGGGGAATAT
TGGACAATGGGCGCAAGCCTGATCCAGCAATGCCGCGGGGTAAGAAGGCCT
TTCGGATCGAAAGCCCTTCGACAGGGACGATAATGACGAACTGTATAGTGC
CCCGGTAATTCNNGGC

3 (EU94527)

ATTTGCGGCGANNNNNNNNNNNNNNNNNNNTCTGCCTTCAACNCTGGGN
NNNNNNNNNNAACCGGGGNTAATACCGGATATGAGCCTTCGCGATCNTCC
GCNTNNNGTTTTTCGGCCTGAGTGATCTCCGGCTTCACCTTGTGGTGGGTA
AGGCTCCCAAGGCACGCCCCGACCCGCCTGGAGGGGACGNCCCCCGGGGC
TGAGACACGCCAATCCCTACGGAGGCACCGTGGGGAAAATGGGNAATGA
GGAACTTGACCCACCACCCCTTGCGCATGAGGCCTTGGGTTTTAACCCCT
TCTTAGGTATTTAGCGCAATAAGGTACCTCCGAAGAGGAGGAGGTNACTAT
TTCCACCGCGCGCTAAAAA

4 (EU94528)

GGTGAGCATGTATTACCGCGGCTGATGTCCCAAAGGCTTAAGNACTAACGC
GGCAGAAGGCCTTCAGGCTGGCGCGGTANGGCAGGATTAGGCTTGGCTNCA
TTGCGTAAAATTCCCCACTGCTGTCTCCCGTANGAGCGGGGAGTGTCTCGCA
GACCATCTACCGGTCCGTCTCTCAGACCAGCTGGACCTCGCAACTATGTTA
TCCCTTTACCCCACTAACTACCTAATCTGACATCGTTNGCCCAACAGCACT
AGGCCTTATGGTCCCCGCTTTTACACGTAGTTCGTATGCGGTATTACTCCG
GTTCTCGCCGCGCTATCCCCACTGTTGCGCACGTTNCGATGCATTACTCAC
CCGTTTTTNACTCGCCGCCGGGTTGNCCCTTGAGTACGGTGGGGCTTGTCAG
TGTAATGCATGCCGCCAGCGTTCAACCTGAGCAAGGATCAAACCTCTCAGA

5 (EU94529)

TGCACGTCGAGCGGCAGCGNGAAAGTAGCTTGCTACTTTTGCCGGGAGNGG
CGGACGGGTGAGTAATGCCTGGGGATCTGCCAGNNGAGGGGGATAACTAC
TGGAACGGTAGCTAATACCGCATAACGCCCTACGGGGGAAAGCAGGGGAC
CTTCGGGCCTTGC GCGATTGGATGAACCCAGGTGGGATTAGCTAGTTGGTG
AGGTAATGGCTACCAAGGCGACGATCCCTAGCTGGTCTGAGAGGATGATC
AGCCACACTGGA ACTGAGACACGGTCCAGACTCCTACGGGAGGCAGCAGTG
GGGAATATTGCACAATGGGGGAAACCCTGATGCAGCCATGCCGCGTGTGTG

AAGAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGAGGAGGAAAGGTTGGTA
GCTAATAACTGCCAGCTGTGACGTTACTCGCAGAAGAAGCACCGGCTAACT
CCGTGCCAGCAGCCNGCGGTAA

6 (EU94530)

TTAGCATGTATTACAGCGACTGCTGTTCCAANGGAGTAGNNCTTCCCCGGC
GGTGCGNCATCGGGNTGGGGTTGATNGNTTTGGACNANATTCNNCACTGTT
GCGTACCATAGTGGTCTGGGCCGTATCTCAGGTGNNGTGNGTCTTCTCTCC
TCTCAGGTCCGCTACCCGNCGNTGCCATGGTGTGGCGTTACCACCCAAACTA
NCTGATAGGCCGCGATCCCATCCTAAACCGAAATTTTTTCCCCACCCNAAGA
TGCCCTAAAGGTTCTGTATCTGGNATTAGGTCCCGTTACCCGGAGTTATCCCC
AAGTGCAGGGCAGATTGCTCACGTGTAACCCACCCGTACCCCACTAATTTGC
CCGGATTTTGCTCCNNNTTCGTCGTTTCGCTGGGGTGTGGTTGGGGGGCCCCA
NCAGCGTTCGTCCTGAGCCAGGATCANACACTCAA

7 (EU94531)

GCTCGGCGGCGTGCCTAACACATGCAAGTCGAACGGGCATCTTCGGTGCGG
GGGGCGGGGGGGTGTGAGTCACGCGAAAGAGTCTTCCTTCGCGGCAGGAACCA
CTGTTGGTAGCGACTGCACATACCCTGTATGTCGGAGGGAGGAACCTAATC
GGCCTAGAGACGCCCTGGCGTCTGATCGACTTGTGGGGGGGAAAGAGCCT
ACCAAGGCCACGATTAATAGGTGGTCTAAGAGGATGAGCAG

8 (EU94532)

GCCTAACACATGCAAGTCGAACGGGAATCTGCGGCAATGGTGGCGGAGGG
GTGACTAACGGGTAAAAATCTAGCGTCGGGACCCGTCCTGCGGTATGTAGC
GATAGCTACTACCCTTTTCTTCGTAAATGGCATGTATTAGCTGTGAAAGGGC
TGCGCTCTGAT

9 (EU94533)

TAACACATGCAAGTCGAACGATAAAATTGTTTTCGAGGGTCAGAGGTGATG
ACGGACGTGAAAGCTATTGGTCTCCCCAGTAACAAGTCTTTAAAGAGATAT
TGAAAAGCCAATAAGACTGTA

10 (EU94534)

GGCGGCGTGCCTAACACATGCAAGTCGAACGGTAATGTGGGTAAACAGCGG
CGGAGGGGTGAGTAGGGGGAAAGAGTAGAAATACGGGCGGGGTGGTGGG
TTAGTAACCGGTGAAAAA

11 (EU94535)

TCGANCGGGAGTATTCGGNTTCTCGTGGCAGANGGGTGNNNNNNNNNNNN
NNNNTNNCTTCANNTCCGGNATNCNGTTGGAAACAAGAGCAANTCCCNAT
ATNCCGCNAGGCGAAACCTAATTGCNCTGGCGAAGAGCTTGTCTGTATNT
TCAGTTGGGGGGNTAAGACCTTACCAAGGCNACTATCAGAAGCTGGNCTGA
GAGGATGAGCAGCCACACTGGGACTGAGACACGGCCCACACTCCTACGGGA
GCCAGCANTGGGAATTTTCCCAATGGGGGAAACCCTGACGGANCAACGC
CGCGGGAGGGAGGAAGGCCTTTGGGTGGAAACCTCTTTTCTCAGGGAAGA
AGTTCTGNCNNTCCTTGATGGATTATCCTCGGNTAACTCCGTGCCAGCCNGC
CGGCGGNAATAGGGGCAAACCACCCCCCANANNCCGNTGCACCCCGCCC
CGGGGAATANANAGAGANNNGGNGACNANNCCN

12 (EU94536)

ACGGNATCTTCGTATTCTAGTGGCGGACGGGTGANTNNNNNNNNNNNGTCTN
NCTTCNGGACNTGNNCCNCGGTTGAAAACANGGGCAACTACCCGATATGCC
GCAAGGTGAAACCTAATTGGCCTGAAGAAGAGCTTGCGTCTGATTTTTTAGT
TGGTGGGGTAAGAGCCTACCAAGGCGACGATCAGTGGCTGGCCTGAGAGGA
TGAGCAGCCCCCTGGGACTGAGACACGGCCCACACTCCTACGGGAGGAAG
CNGTGGGGAATTTTCCGCAATGGGCGAAAGCNTGACGGAGCAACGCCGCGT
GAGGGAGGAAGGNCTTTGGATTGTAAACCTCTTTTCTCAAGGAAGAAGTTC
TGACGGTACTTTGAGGAATTTGCCTCGGCTAACTCCGTGCCAGCAGCCGCGG
GAATACNTGCAA

13 (EU94537)

TGGTGAGCAT GTATTACAGC GGCTGCTGGC CAAAGGTGAG TNNNANTACC
GCGGCTGTTGGTCTCGAGGNTTCTCTTTTGCGAAAATTCCCTACTGGTGT
CGTCGTAATTCTTGGTCCGTCTCTCAGTCCCAGTGTGGGTGATCATCCTCTCA
GAAGGTGTACTGCTCTTCGCCGTGATGAGCTTTTACCCCTGCTATGTGATA
ACCTGACGCCAGCCTCNATTTTACCGGANNTCTCTTTCCCCACAGCATATT

GGTATTAAGCAATTTTCCAACCTGGTGTCTCCGCCGNCAAGATAAAATTTCA
CGCGGGNNCCCCCCCCCCCCCAATAAAATACGAANATCTTGNTACAACCTG
AATGAATGAGTCACTCCGGCGTGTTTCATCCGGAGCCAGGANAAATCCTCG
AAAGAGGGNCTCNNGCTCACATCN

14 (EU94538)

TGGTGAGCCCGTATTACCGCGACTGCTGGCCNAAAGNCTTNNNNNNNACG
CGGCAGTTGTGCCTCAGGGTTTCTTCCATNGNGCAAATTTCCCACTGGTGC
CTCCCGTAGGAGTGCGGGCCGTGGCTCAGNCCCANTGGGGNTGGCCATTCT
CTTAAACCAACTAACGGTCATCGCCATGGTAGGCCCTTGTCCGACCANCTAG
CTAATCATACGCACGCTCTTCTTACCCCAACAAATCTTTCATGCTAAACGTC
ATATTCTAGCACCTATGCGGTATCCGAACGGGGTTCCAGATGTGATCCCCA
GTGTAAGGGAGATTACCCCCGCGTTACTCACCCATCCGAAAATGATGNATC
TCCGAAGATACCTTATTGACCCACTTGGATGTCTTCGGCGGTC

15 (EU94539)

GCCTAACACATGCAAGTCGAACGGTAAAGTGGGTTAGAGAGTGTTCTGGGG
GCGAACGGGGGCGAATCTGTTACGACACTCCCTTCTACACAGGGAAAGCAT
TGGGAAACCGGTGCTAATCCCGCATATTGAAGCTTAATTGACATGGGGAAC
ATCTATTCAAAGAAAAGTGAATTAGTTTCAAACGCCCAAC

16 (EU94540)

GCTTAATACATGCAAGTCGAACGGGAAAGTTGGCAGAGAGGGATGAGGGC
GCTGGATGGGACGATCTGTGTCGACCATCCCTTTCGTACAGTGAAAGAGGC
GCGAAAACGGTATAAACACTTAATGTTAAAGATTAATGCCATAAAAGACG
TGAGTATAT

17 (EU94541)

GCTCGAACGCTCGGCCGGCAGGCCTAACACATGCAAGTCGAACGGAAAGCTT
ACAGAGTGGGTGACGGGTGAGTAACATGCGGGAATCCGCCTTGTGGTTCCG
GTCAACATTGGGATACCGGTGCTAAAACCTAGATAAATCCTCACGGGGAAAG
TTTTAATGCCATAAGATGAGCCCGGATTTCGATTAGTTAGTTGGGGAGGGAA
AGACTCTCCAAGACAATGATTAATAGCTGATCTGAGAGGATGAACC

18 (EU94542)

GCCTAACACATGCAAGTCGAACGGTAATGTTTCGTATGCTAGCGGCGGACGG
GTGAGTAACGTGTAAGAATCTATCTTCACTACGTTTACAACGGTTGGAAACG
ACAGCAAATACTCGATATGCCGCAAGGTGAAACCTAATTGGCCTGGAGAAC
AGCTTGCGTCTGATTA GCTAGTTGGGGGGGTAA

2. Sequence of unknown bacteria (Chapter 6), 100% match to *Bacillus cereus*

GCCAGCTTATTCAACTAGCACTTGTTCTTCCCTAACCAACCGATAATTACGAC
CCGAAAGCCTTCATCACTCACGCGGCGTTGCTCCGTCAGACTTTCGTCCATT
GCGGAAGATTCCCTACTGCTGCCTCCCGTAGGAGTCTGGGCCGTGTCTCAGT
CCCAGTGTGGCCGATCACCTCTCAGGTCGGCTACGCATCGTTGCCTTGGTG
AGCCGTTACCTCACCAACTAGCTAATGCGACGCGGGTCCATCCATAAGTGA
CAGCCGAAGCCGCCTTCAATTTTGAACCATGCGGTTCAAATGTTATCCGG
TATTAGCCCCGGTTTCCCGGAGTTATCCAGTCTTATGGGCAGGTTACCCAC
GTGTTACTCACCCGTCCGCCGCTAACTTCATAAGAGCAAGCTCTTAATCCAT
TCGCTCGACTTGCATGTATTAGGCACGCCGCCAGCGTTCATCCTGAGCCAGG
ATCAAACCTCTC

Appendix B

Presentations and Publications Arising From This Research

G. Ross & T.E. Cloete, 2006. The control of cyanobacterial blooms using predatory bacteria and Phoslock. The 14th Biennial Congress of the South African Society for Microbiology, 9-12 April 2006.

G. Ross & T.E. Cloete, 2006. The use of Phoslock[®] for the control of eutrophication. IWA International Conference, Beijing, September 2006.

T.E. Cloete & G. Ross, 2006. The control of cyanobacterial blooms using predatory bacteria and Phoslock[®]. International Conference and Exhibition on Water in the Environment. 20-22 February 2006, Stellenbosch, South Africa.

Gumbo J.R., G. Ross & T.E. Cloete, 2007. The biological control of *Microcystis* dominated harmful algal blooms. Submitted to *Harmful Algae*.

G. Ross, F. Haghseracht & T.E. Cloete, 2008. The effect of pH and anoxia on the performance of Phoslock[®], a phosphorus binding clay. *Harmful Algae*. 7(4):545-550.

G. Ross, A.K.J. Surridge & T.E. Cloete, 2008. Analysis of the microbial community diversity in Phoslock[®] treated and control areas of Hartbeespoort Dam using PCR-denaturing gradient gel electrophoresis. Submitted to *Water Research*.

G. Ross J.R. Gumbo & T.E. Cloete, 2008. The mechanism of *Microcystis aeruginosa* cell death upon exposure to *Bacillus mycoides*. IWA International Conference, Vienna, September 2008.