

List of Figures

Figure 1. 1 Schematic diagram of biofilm reactors.....	13
Figure 1.2 Steps involved in biofilm formation.....	24
Figure 2.1 Standard graphs for (a) nitrate (b) nitrite and (c) ammonia estimation.....	42
Figure 2.2 Changes observed in appearance of Winogradsky columns spiked with nitrate and activated sludge from three different sources.....	44
Figure 2.3 Amyloid producing bacteria showing red colonies on CPNA plates.....	46
Figure 2.4 Gas production in PNB by the amyloid producing isolates.....	47
Figure 2.5 (a) Denitrification efficiency and (b) Biofilm forming ability of selected amyloid producing denitrifying bacterial isolates.....	49
Figure 2.6 Nitrite accumulation test of amyloid producing denitrifying isolates.....	49
Figure 2.7 Neighbor joining phylogenetic tree depicting phylogenetic relationship of the selected five isolates on the basis of 16S rDNA sequences.....	51
Figure 2.8 Surface amyloid staining with Thioflavin T of the individual isolates of consortium DC5 viz. isolates R4, V5, V9, V11 and V14	52
Figure 2.9 Growth profiles of the selected five isolates of consortium DC5.	53
Figure 2.10 The selected five denitrifying isolates showing lack of inhibition by each other in co-culture growth inhibition assay	53
Figure 2.11 Flask level denitrification studies using selected isolates of consortium DC5..	55
Figure 2.12 Gas chromatographs of N ₂ gas produced by the 5 isolates and consortium DC5	62
Figure 2.13 Kinetic model plots for the removal of nitrate by denitrifying isolates	64
Figure 2.14 Effect of metal ions biofilm formation of consortium DC5.	66
Figure 2.15 Effect of metal ions on denitrification efficiency of consortium DC5.....	67
Figure 2.16 Effect of pH on (a) Denitrification efficiency and (b) Biofilm formation of consortium DC5.....	68
Figure 2.17 Effect of different carbon sources on (a) Denitrification efficiency (b) Biofilm formation of consortium DC5.	70

Figure 2.18 Semi-quantitative PCR analysis showing amplicons of individual isolates of consortium DC5 when grown together in different carbon sources	70
Figure 2.19 Effect of different inoculum (%) on (a) Denitrification efficiency (b) Biofilm formation of consortium DC5.....	71
Figure 3.1 Experimental setup of dMBBR	76
Figure 3.2 Bench scale dMBBR setup developed with consortium DC5 used in denitrification studies.....	81
Figure 3.3 Performance of dMBBR inoculated with consortium DC5 at different (a) C/N ratio, (b) HRT and (c) Nitrate loading	84
Figure 3.4 Carriers with different design and surface area used for dMBBR studies	85
Figure 3.5 dMBBR studies with different carrier design and filling ratio.....	87
Figure 3.6 Stover-Kincannon model for denitrifying MBBR (a) Predicted, (b) Observed model.....	90
Figure 3.7 Comparative studies between suspended growth reactor and dMBBR.....	91
Figure 3.8 Comparative studies between MBBR developed with consortium DC5 and MBBR developed with activated sludge.....	93
Figure 3.9 Comparative studies between dMBBR developed with consortium DC5 and control dMBBR.	94
Figure 3.10 (a) Nitrate and (b) COD removal studies from dye industry effluent.	96
Figure 3.11 (a) Nitrate and (b) COD removal studies from pharma industry 1 effluent. .	97
Figure 3.12 (a) Nitrate and (b) COD removal studies from pharma industry 2 effluent..	97
Figure 3.13 (a) Nitrate and (b) COD removal studies from pharma industry 3 effluent..	98
Figure 3.14 (a) Nitrate and (b) COD removal studies from domestic wastewater spiked with nitrate.	98
Figure 4.1 Abundance of different phyla in denitrifying biofilm developed with consortium DC5	106
Figure 4.2 Abundance of different genera in denitrifying biofilm developed with consortium DC5	107
Figure 4.3 Abundance of different <i>Thauera</i> spp. and <i>Paracoccus</i> spp. in denitrifying biofilm developed with consortium DC5.....	107
Figure 4.4 PCA component analysis (a) Phyla level (b) Genera level (c) Species level	109

Figure 4.5 Relative abundance of genes of metabolic pathways in denitrifying biofilm developed with consortium DC5 (a) KO level 1 (b) KO level 2	111
Figure 4.6 Relative abundance of genes resistance to heavy metal, antibiotic and toxic compounds in biofilm developed by consortium DC5	113
Figure 4.7 Different strategies of anaerobic/anoxic biodegradation of organic compounds in bacteria.....	114
Figure 4.8 (a) Relative abundance of gene involved in nitrogen metabolism (b) Nitrogen metabolism pathways by KEGG mapper in biofilm associated with carriers in dMBBR	117
Figure 4.9 (a) Relative abundance of genes involved in sulfur metabolism (b) Sulfur metabolism pathways by KEGG mapper.....	120
Figure 4.10 (a) Relative abundance of genes involved in methane metabolism (b) Methane metabolism pathways by KEGG mapper in biofilm associated with carriers in dMBBR.....	123
Figure 4.11 Relative abundance of genes involved in xenobiotic biodegradation and metabolism by KO	125
Figure 5.1 (a) Auto aggregation ability (b) Hydrophobicity of <i>Thauera</i> sp.V14	137
Figure 5.2 Effect of temperature on morphology and swarming motility of <i>Thauera</i> sp.V14(a) 25 °C, (b) 30 °C ,(c) 37 °C, (d) 40 °C.	138
Figure 5.3 Flask level denitrification studies with <i>Thauera</i> sp.V14.....	139
Figure 5.4 Effect of inoculum on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 (b) Biofilm of <i>Thauera</i> sp. V14.....	140
Figure 5.5 Effect of metal ions on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 in presence of MgCl ₂ (b) Biofilm of <i>Thauera</i> sp. V14 in presence of MgCl ₂ (c) Denitrification efficiency of <i>Thauera</i> sp.V14 in presence of CaCl ₂ (d) Biofilm of <i>Thauera</i> sp. V14 in presence of CaCl ₂	141
Figure 5.6 Effect of Purified amyloid protein on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 (b) Biofilm of <i>Thauera</i> sp. V14.....	142
Figure 5.7 Effect of NaCl on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 (b) Biofilm of <i>Thauera</i> sp. V14.	143

Figure 5.8 Effect of DMSO on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 (b) Biofilm of <i>Thauera</i> sp. V14.....	144
Figure 5.9 Effect of Ethanol on (a) Denitrification efficiency of <i>Thauera</i> sp.V14 (b) Biofilm of <i>Thauera</i> sp. V14.....	145
Figure 5.10 Contour and 3-D graphs of (a) Effect of Yeast extract and Peptone (b) Effect of Ethanol and DMSO (c) Effect of CaCl ₂ and Peptone on biofilm forming ability of <i>Thauera</i> sp.V14.....	151
Figure 5.11 Contour and 3-D graphs of (a) Effect of Peptone and CaCl ₂ (b) Effect of Ethanol and DMSO (c) Effect of Peptone and Yeast extract on denitrification ability of <i>Thauera</i> sp.V14.....	154
Figure 5.12 Validation of RSM results at flask level (a) Denitrification efficiency of <i>Thauera</i> V.14, (b) Biofilm forming ability of <i>Thauera</i> sp.V14.....	155
Figure 5.13 Nitrate removal studies in continuous dMBBR inoculated with <i>Thauera</i> sp.V14.....	156
Figure 5.14 (a) FTIR of the biofilm developed on the carrier, (b) SEM image of the surface of the carrier before and after biofilm development in dMBBR developed with <i>Thauera</i> sp.V14.....	157