**Table S1.** Primer sequences used in the study and the amplicon sizes of target genes.

| Target strain | Primer Sequence (5′→3′) | Reaction Conditions (ºC) | Cycles | Amplicon size | Reference |
|---|---|---|---|---|---|
| *E. coli* | UAL754: 5′-AAAACGGCAAGAAAAAGCAG-3′<br>UAR900: 5′-ACGCGTGGTTACAGTCTTGCG-3′ | 94°, 94°, 60°, 72°, 72°<br>2′, 90′′, 90′′, 90′′, 10′ | 35 | 147 bp | [1] |
| *Vibrio* spp. | V.16S-700F: 5′-CGGTGAAATGCGTAGAGAT-3′<br>V.16S-1325R: 5′-TTACTAGCGATTCCGAGTTC-3′ | 93°, 92°, 57°, 72°, 72°<br>15′, 40′′, 60′′, 90′′, 7′ | 35 | 663 bp | [2] |
| *blaTEM* | temF: 5′-TTTCGTGTCGCCCTTATTCC-3′<br>temR: 5′-CCGGCTCCAGATTTATCAGC-3′ | 94°, 94°, 60°, 72°, 72°<br>5′, 30′′, 30′′, 90′′, 5′ | 30 | 690 bp | [3] |
| *blaSHV* | shvF: 5′-AGGATTGACTGCCTTTTTG-3′<br>shvR: 5′-ATTTGCTGATTTCGCTCG-3′ | 94°, 94°, 54°, 72°, 72°<br>5′, 30′′, 30′′, 1′, 10′ | 32 | 393 bp | [3] |
| *blaZ* | zF: 5′-ACTTCAACACCTGCTGCTTTC-3′<br>zR: 5′-TGACCACTTTTATCAGCAACC-3′ | 94°, 94°, 60°, 72°, 72°<br>5′, 30′′, 30′′, 90′′, 5′ | 30 | 490 bp | [4] |
| *blaCTX-M* | ctxF: 5′-CGATGTGCAGTACCAGTAA-3′<br>ctxR: 5′-TTAGTGACCAGAATAAGCGG-3′ | 94°, 94°, 60°, 72°, 72°<br>5′, 30′′, 30′′, 90′′, 5′ | 30 | 585 bp | [3] |
| *aadA* | aadF: 5′-GTGGATGGCGGCCTGAAGCC-3′<br>aadR: 5′-AATGCCCAGTCGGCAGCG-3′ | 94°, 94°, 68°, 72°, 72°<br>4′, 45′′, 45′′, 45′′, 7′ | 30 | 525 bp | [5] |
| *strA* | strF: 5′-CTTGGTGATAACGGCAATTC-3′<br>strR: 5′-CCAATCGCAGATAGAAGGC-3′ | 94°, 94°, 68°, 72°, 72°<br>4′, 45′′, 45′′, 45′′, 7′ | 30 | 546 bp | [5] |
| *tetA* | aF: 5′-GCTACATCCTGCTTGCCTTC -3′<br>aR: 5′-CATAGATCGCCGTGAAGAGG-3′ | 94°, 94°, 55°, 72°, 72°<br>5′, 60′′, 60′′, 90′′, 5′ | 35 | 210 bp | [6] |
| *tetB* | bF: 5′-TTGGTTAGGGGCAAGTTTTG-3′<br>aR: 5′-GTAATGGGCCAATAACACCG-3′ | 94°, 94°, 55°, 72°, 72°<br>5′, 60′′, 60′′, 90′′, 5′ | 35 | 659 bp | [6] |
| *tetK* | kF: 5′-GTAGCGACAATAGGTAATAGT-3′<br>kR: 5′-GTAGTGACAATAAACCTCCTA-3′ | 94°, 94°, 55°, 72°, 72°<br>5′, 60′′, 60′′, 90′′, 5′ | 35 | 460 bp | [6] |
| *tetM* | mF: 5′-AGTGGAGCGATTACAGAA-3′<br>mR: 5′-CATATGTCCTGGCGTGTCTA-3′ | 94°, 94°, 55°, 72°, 72°<br>5′, 60′′, 60′′, 90′′, 5′ | 35 | 158 bp | [6] |

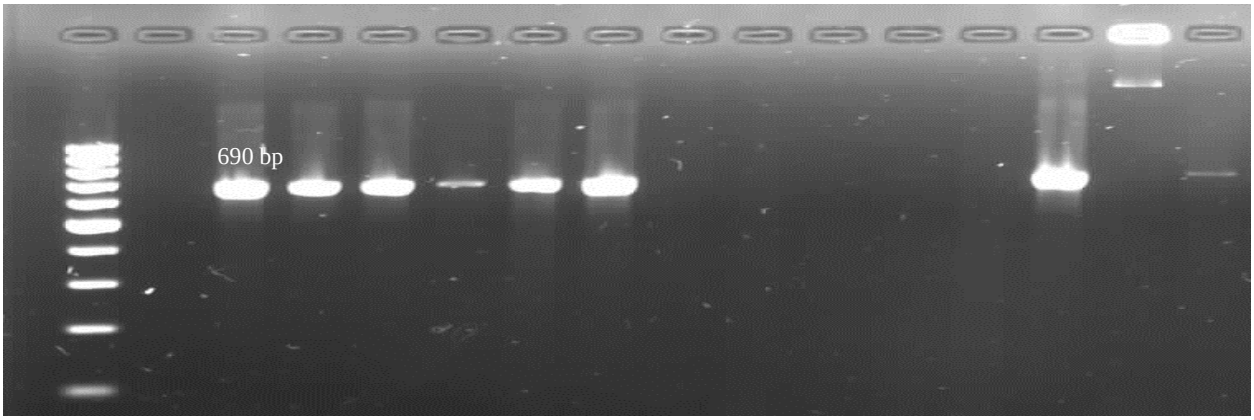
**Table S2.** Regulatory limits for discharged effluents into water resource.

| Parameter (Unit) | Regulatory guideline | | Reference |
|---|---|---|---|
| | **General limit** | **Special limit#** | |
| Faecal Coliforms | 1000 CFU/ 100 ml | 0 | [7] |
| pH | 5.5 – 9.5 | 5.5 – 7.5 | |
| Electrical Conductivity (mS/cm) | 70 mS/cm above intake to a maximum of 150 mS/cm | 70 mS/cm above background receiving water, to a maximum of 100 mS/cm | |
| Free Chlorine (mg/l) | 0.25 mg/l | 0 | |
| Nitrate ($NO_3^-$) (mg/l) | 15 mg/l | 1.5 | |
| Nitrite ($NO_2^-$) (mg/l) | 15 mg/l | 1.5 | |
| Phosphate ($PO_4^-$) (mg/l) | 10 mg/l | 1 (median) to 2.5(maximum) | |
| Chemical Oxygen Demand (COD) (mg/l) | 75 mg/l after removal of algae | 30 mg/L after removal of algae | |
| Temperature ($^oC$) | Maximum of 35 °C | Maximum of 25 °C | [8] |
| Suspended Solids (mg/l) | Not exceed 90 mg/l | Not exceed 10 mg/l | |
| Colour, Odour or Taste | Nil | Nil | |
| Soap, Oil and Grease | Not exceed 2.5 mg/l | Nil | |
| Total Dissolved Solid (TDS) (mg/l) | 450 mg/l | - | [9] |
| Turbidity (NTU) | <5 NTU | - | [10]* |
| Biochemical Oxygen Demand ($BOD_5$) (mg/l) | 3 – 6 | - | [11]* |
| Dissolved Oxygen (mg/l) | ≥ 5 | - | |

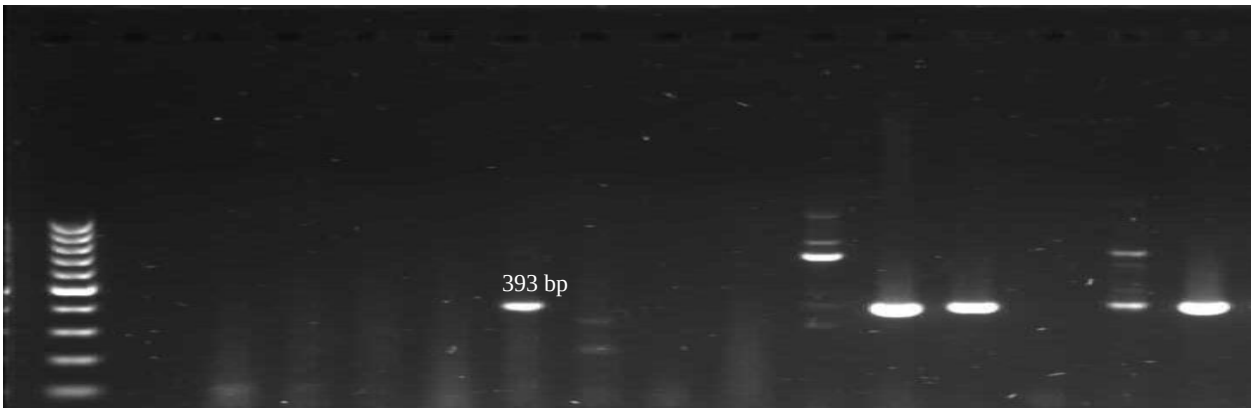
*International guidelines were used where limit does exist in South African guidelines.

#Special limits, as defined by the Department of Water and Sanitation (formerly Department of Water Affairs and Forestry), are often enforced where more stringent effluent quality control is required due to the sensitive nature of some catchment areas.

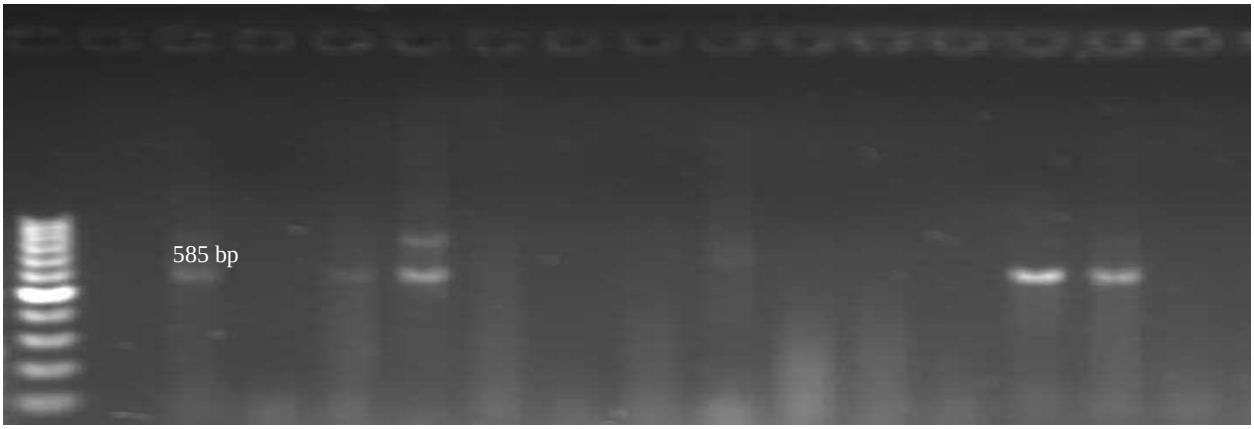
**Figure S1: Gel electrophoretic pictures showing the resistance genes detected in the vibrio isolates.**



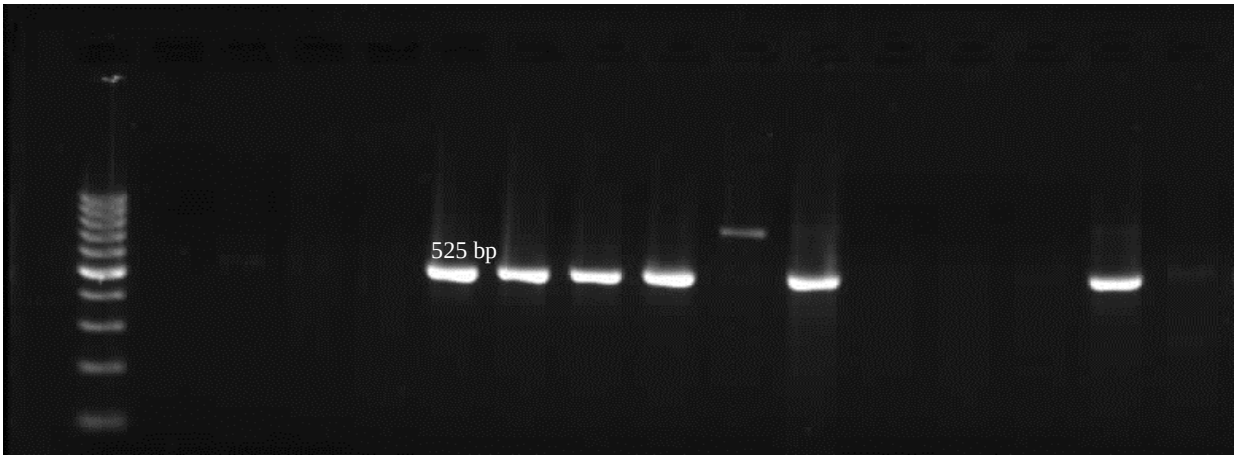

(a) Molecular detection of *bla*TEM gene (690 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.



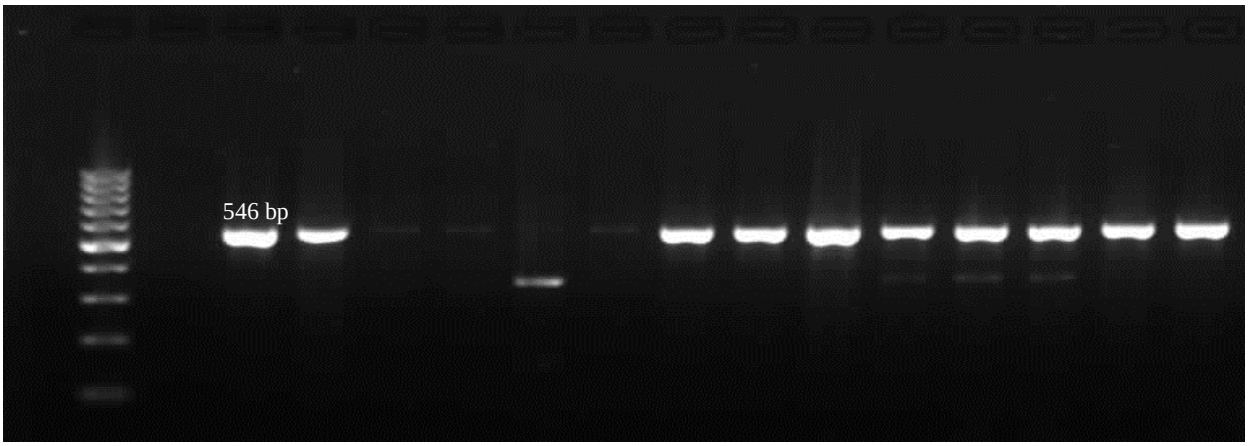

(b) Molecular detection of *bla*SHV gene (393 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.



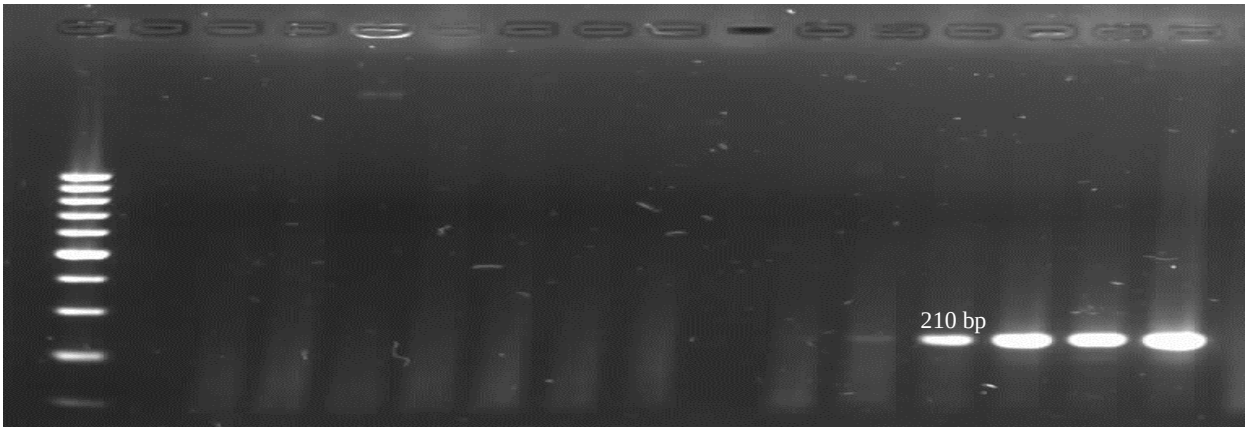

(c) Molecular detection of *bla*CTX-M gene (585 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.



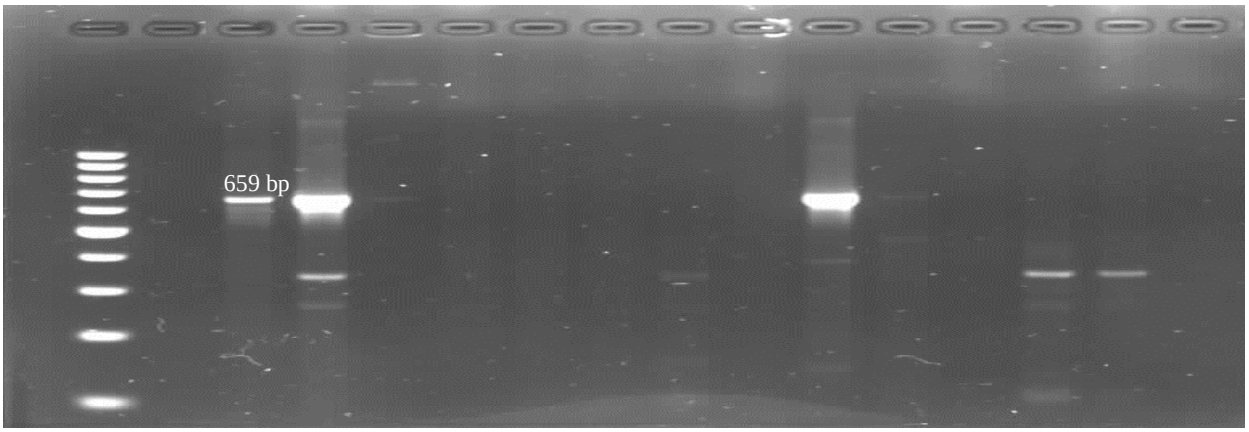

(d) Molecular detection of *aadA* gene (525 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.



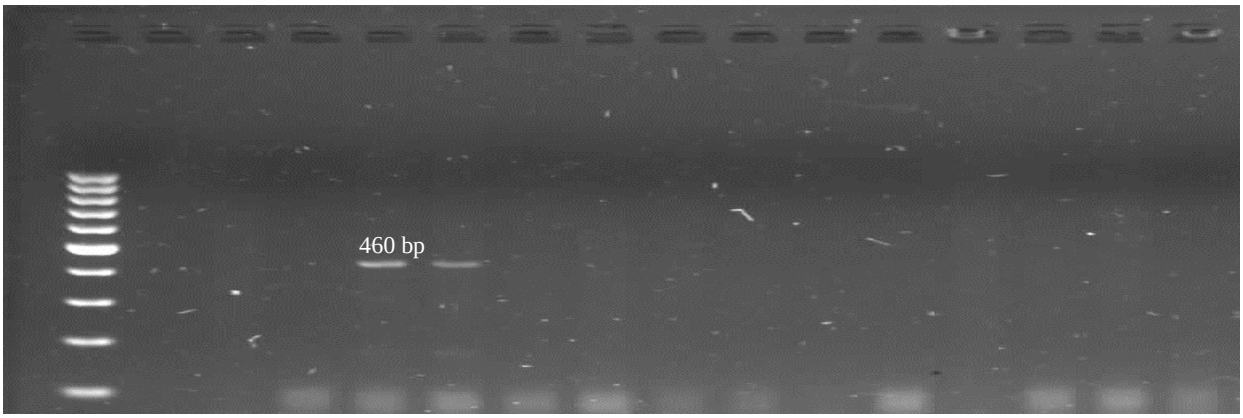

(e) Molecular detection of *strA* gene (546 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.



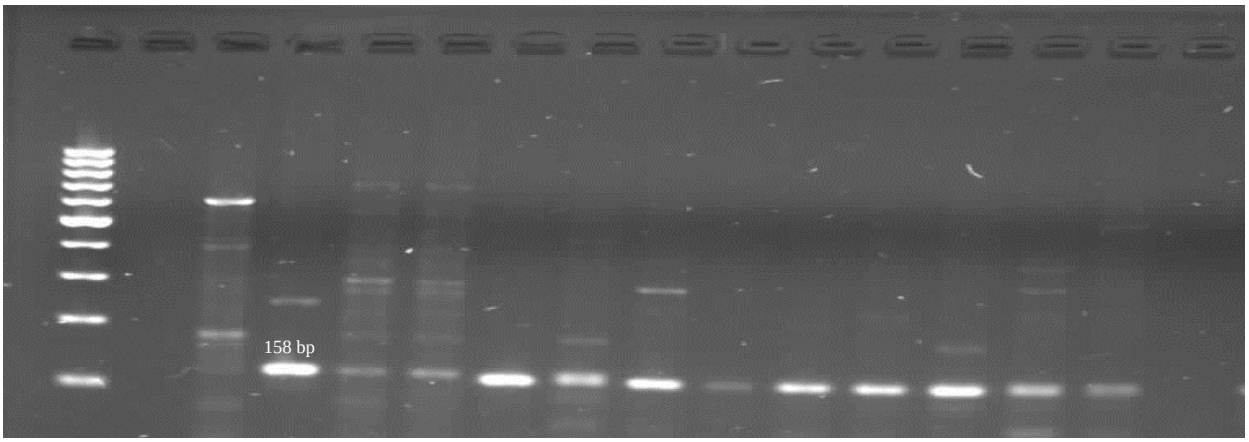

(f) Molecular detection of *tetA* gene (210 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.




(g) Molecular detection of *tetB* gene (659 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.




(h) Molecular detection of *tetK* gene (460 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.




(i) Molecular detection of *tetM* gene (158 bp). Legend: Lane 1: 100 bp Molecular weight marker; lane 2: Negative control; lanes 3 to 16 *Vibrio* isolates.

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