Table S4. Successful PCR conditions for the amplification of *msmA* and *msmE* gene sequences from SCD0 and SCDE samples.

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| --- | --- | --- | --- | --- | --- |
|  | **Nested PCR for amplification of *msmA* sequences** | | | **Nested PCR for amplification of *msmE* sequences** | |
| **Primers SarA124fwd/SarA1053rev**  **(First PCR)** | | **Primers SarA139fwd/SarA488rev (Second PCR)** | **Primers SarE133fwd/SarE1125rev**  **(First PCR)** | **Primers SarE322fwd/SarE704rev**  **(Second PCR)** |
| **Metagenomea** | SCD0 | SCDE | SCD0 and SCDE | SCD0 and SCDE | SCD0 and SCDE |
| **PCR program** | 95oC 2 min + 35 x (94oC 1 min + 56.2oC 1 min + 72oC 1min) + 72oC 5 min | 95oC 2 min + 35 x (94oC 1 min + 52.1oC 1 min + 72oC 1min) + 72oC 5 min | 95oC 2 min + 30 x (94oC 1 min + 51oC 30 seg + 72oC 1min) + 72oC 5 min | 95oC 2 min + 35 x (94oC 1 min + 58.2oC 1 min + 72oC 1min) + 72oC 5 min | 95oC 2 min + 30 x (94oC 1 min + 55oC 30 seg + 72oC 1min) + 72oC 5 min |
| **Concentrationsb** | 0.8 µM of forward and reverse primers, 1.25 U of GoTaq® G2 Flexi DNA polymerase (Promega). | 0.8 µM of forward and reverse primers, 1.25 U of GoTaq® G2 Flexi DNA polymerase (Promega), with 0.125 M betaine and 2.5% DMSO | 0.8 µM of forward and reverse primers, 1.25 U of GoTaq® G2 Flexi DNA polymerase (Promega). | 0.8 µM of forward and reverse primers, 1.25 U of GoTaq® G2 Flexi DNA polymerase (Promega). | 0.8 µM of forward and reverse primers, 1.25 U of GoTaq® G2 Flexi DNA polymerase (Promega). |

(a) The DNA was first amplified using REPLI-g® MiniKit (QIAGEN)

(b) In all cases, PCR was performed in a 25 L volume using the manufacturer’s buffer associated with the Taq polymerase employed, 1.5 mM MgCl2 and 200 µM of each dNTP.