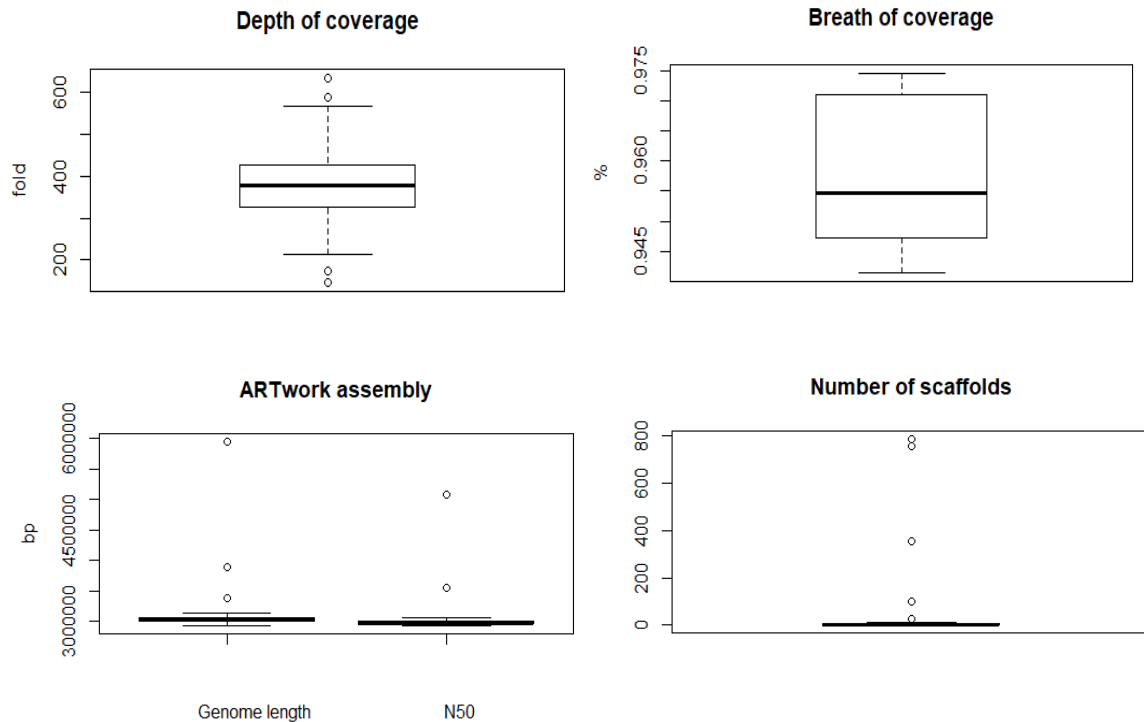


1 **Additional file 4. Quality metrics of mapping (i.e. iVARcall2) and *de novo* assembly (i.e. ARTwork)**
2 **from the studied *Listeria monocytogenes* genomes**

3 An essential step in comparative genomics based studies is the Quality Control (QC) of WGS data in
4 order to guarantee the accuracy of sequencing results obtained by *in silico* genome-wide analysis. Poor
5 quality of read sequences as well as contamination of DNA can lead to significant errors in variant
6 calling (low-depth of sequencing impact on false-positive rate) and gene prediction analyses [1]. Even
7 though the harmonization and standardization of WGS data analysis is still an ongoing process [2], a
8 number of metrics for QC of *de novo* draft genome (i.e. contiguity of assemblies) and genome coverage
9 (i.e. number of reads mapped to a specific position within the reference genome, so called
10 “mappability”) is currently available [3–5]. In this study, standard quality metrics of reads mapping (i.e.
11 iVARcall2) and *de novo* assembly (i.e. ARTwork) obtained from Illumina paired end reads of 96 *Listeria*
12 *monocytogenes* genomic DNA were assessed and reported in the boxplots. In particular, the quality of
13 reads mapping onto the reference genome was evaluated based on the depth of coverage (average
14 number of times that a base of a genome is sequenced) and breadth of coverage (percentage of bases
15 of a reference genome that are covered with a certain depth). Moreover, contiguity measures, such as
16 the size of assembled genomes (express in total number of bases and representing an indicator of
17 exogenous DNA contamination), and the total number of contigs/scaffolds along with the N50 value
18 (size of the largest contig, or scaffold, for which half the total size is contained in that contigs and those
19 larger), were calculated. Overall, high values of depth and breadth of coverage (1st and 3rd quartile =
20 145-426X and 94-96%, respectively) have been estimated confirming the high quality of the Illumina
21 short reads for further analyses. Accordingly, genome sizes as well as the number of scaffolds and the
22 N50 (median values of 3,021,803 bp, 4 and 2,969,304 bp, respectively) demonstrated high
23 performance of ARTwork pipeline in term of high contiguity of *de novo* assemblies for almost all *L.*
24 *monocytogenes* samples (~98%). However, two out of 96 assemblies were discarded from further
25 analyses since suspected of contamination (total genome length higher than 3.8 Mbp and number of

26 scaffolds > 354).



27

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