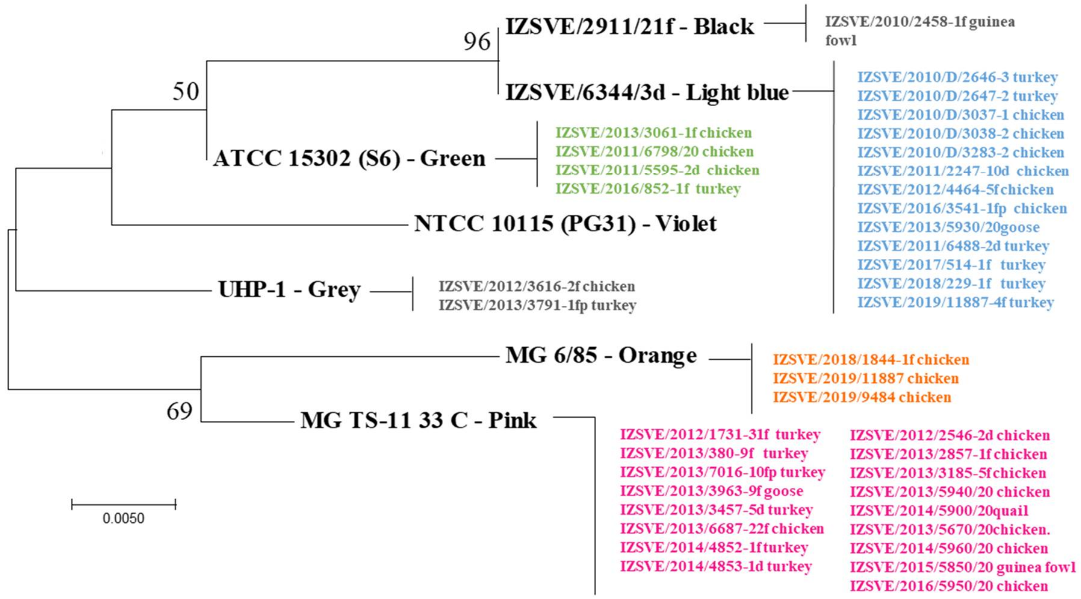

Figure S1. Phylogenetic tree based on the *mgc2* sequence of the MG isolates. Sample name and host are reported in font color accordance with the relative *mgc2* type. Cutoff of bootstrap values was set to 50. Phylogenetic distance bar is represented below.

Table S1. Primer sequences and expected amplicon size (bp) of the PCR protocols described in this study.

Gene Target	Primer Forward (5′->3′)	Primer Reverse (5′->3′)	PCR Amplicon Size (bp)
gapA	GAACTGATTTTGGAAGCGTAAGA	CCGTTATATCATCCTGAATCTCCT	110
mgc2	CGCAATTTGGTCCTAATCCCCAACA	TAAACCCACCTCCAGCTTTATTTC C	237–300
atpG	TGGAACTAAACTAAATTCGTTTTTAAGA	TAGCATACTCACACACTTTGGATTCA	395
dnaA	GAGCGTCAAAAATTATTCCCAGA	TTACGAATATCGCCTTCATCAAA	461
fusA	CAGTAGCAGTATTAGATGCCCAAATG	TAGTAGGGATCTGTACTTCTTCACCAA	597
rpoB	GTTAATGCTTAAAGAACAACTTGATTTATT	GGTTAATTGGTGCGTGTTAAAGAA	562
ruvB	CAACGACAATGTATGGCAGGAT	AAACAATCAATTCCACTTATTAGTGAAA	388
uvrA	TTTACCAATCTTAATGTGAATAAAGCC	CCGTTCCCTGGGTGGAGTT	536

Table S2. Allele distribution into sequence type of Italian samples analyzed.

MLST Target	Observed Number of Allele Variants (Number of Known Variants [1])	Prevalent Allele Number [2] (Number of Samples)
atpG	5 (24)	8 (23). 2 (6)
dnaA	7 (22)	1 (23), 5(6)
fusA	8 (27)	16 (20), 14 (5)
rpoB	12 (33)	15 (11), 3 (4), 5 (4), 7 (4), 8 (4), 17 (4)
ruvB	5 (24)	1 (15), 2 (14)
uvrA	7 (24)	12 (17), 5(8)

[1] Based on https://pubmlst.org/mgallisepticum/ updated on August 2020. [2] Allele sequence present in >50% of samples.

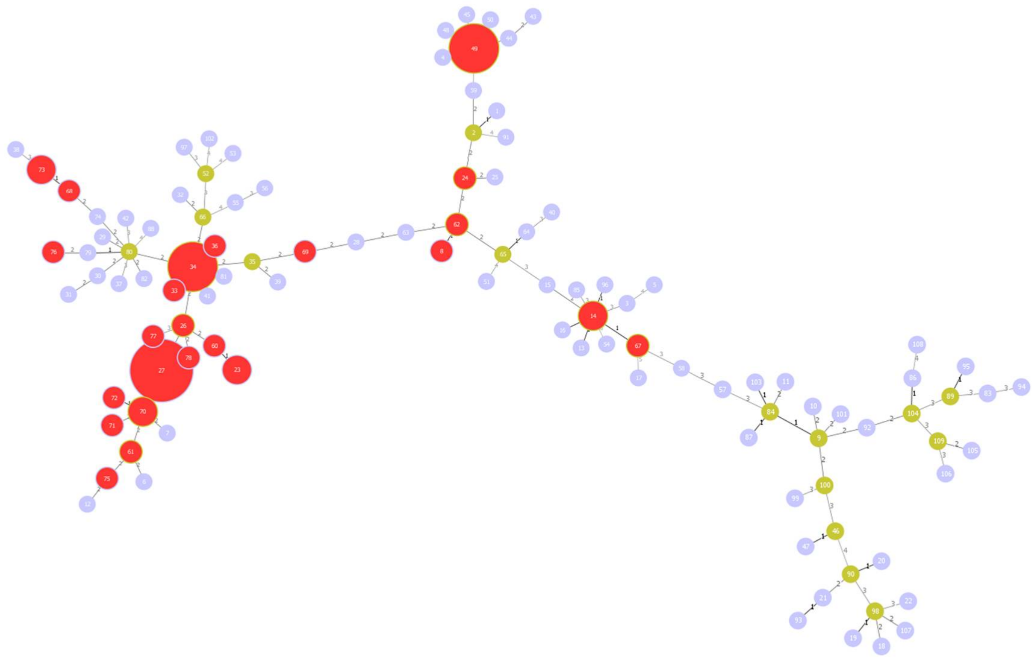

Figure S2. Full MST tree MG MLST 6-genes. Dispersion of full MST of all described six-alleles scheme ST profiles with PubMLST database updated to August 2020. Red circles are Italian ST described in this study, green ST the nodal points, and in light grey, other known STs.