



## Supplementary file

**Table S1:** Minimum inhibitory concentration breakpoints as per the CLSI guidelines

Drugs	Sensitive (S) mg/L	Intermediate (I) mg/L	Resistant (R) mg/L
Metronidazole	≤ 8	16	≥ 32
Clindamycin	≤ 2	4	≥ 8
Imipenem	≤ 4	8	≥ 16
Piperacillin-tazobactam	≤ 16/4	32/4-64/4	≥ 128/4
Chloramphenicol	≤ 8	16	≥ 32
Cefoxitin	≤ 16	32	≥ 64

**Table S2:** PCR primers and the thermal cycling parameters used to detect the target AMR genes and IS elements

Gene	Primer sequence (5'–3')	Size	PCR conditions	Ref.
<i>nim</i>	F-ATGTTTCAGAGAAATGCGGCGTAAGCG R-GCTTCCTTGCCTGTCATGTGCTC	458 bp	94°C for 5 min; 35 cycles of 94°C for 30 sec, 63°C for 55 sec, 72°C for 45 sec.	[3]
<i>cfiA</i>	F-ATG GTACCTTCCAACGGG R-CACGATATTGTCTCGGTCGC	353 bp	94°C for 5 min; 35 cycles of 94°C for 1 min, 52°C for 1 min, 72°C for 1 min.	[54]
<i>IS1186</i>	F-GAGAATCAAGCTTCTCGCC R-CCCCGAATTCGCCTTTGCCCGTA	1-6 kb	94°C for 5 min; 35 cycles of 98°C for 10 sec, 60°C for 30 sec, 68°C for 1 min.	[55,56]
<i>cfiA<sup>IS</sup></i>	G'-CGCCAAGCTTTGCCTGCCATTAT E'-CTTCGAATTCGGCGAGGGATACATAA	<sup>a</sup> 1.6-1.7 kb <sup>b</sup> 350 bp	95°C for 1 min; 35 cycles of 95°C for 20 sec, 64°C for 2 min, 72°C for 1 min.	[57]
<i>cepA</i>	F: TTTCTGCTATGTCCTGCCC R: ATCTTTCACGAAGACGGC	780 bp	98°C 10 s, 60°C 30s, 68°C 1 min, 30 cycles	[3]
<i>cfxA</i>	F: ATCGTAGTTTTGAGTATAGCT R: TAAAAGCACTCCGATAACGAT	1010 bp	94°C 1 min, 56.5°C 45 s, 72°C 2 min, 30x	[3]
<i>ermF</i>	F: CGGGTCAGCACTTTACTATTG R: GGACCTACCTCATAGACAAG	466 bp	94°C 30s, 50°C 30 s, 72°C 2 min, 35x	[3]
<i>cat</i>	F: CCTGCCACTCATCGCAGT R: CCACCGTTGATATATCCC	623 bp	94°C 30s, 54°C 30 s, 72°C 2 min, 35x	[58]

F: forward, R: reverse. *cfiA<sup>IS</sup>*, *cfiA* gene (350 bp), and the intact segment containing *cfiA* gene and upstream insertion sequence elements (1.6-1.7 kb).

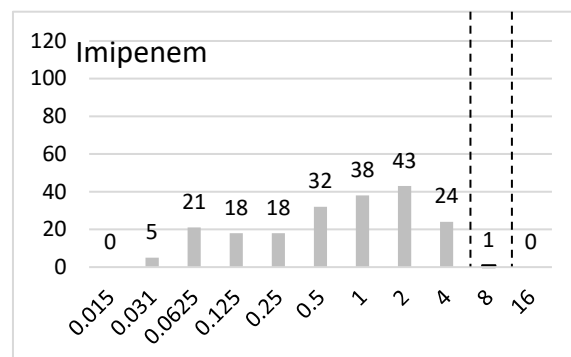
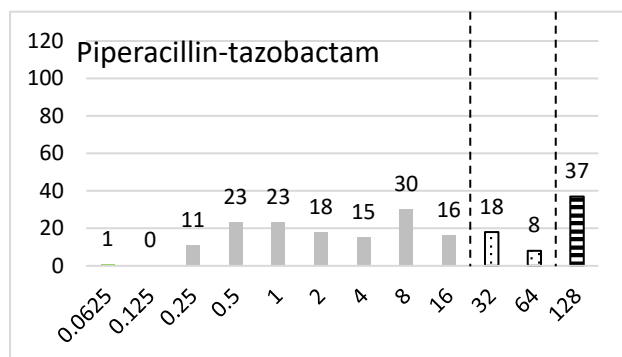
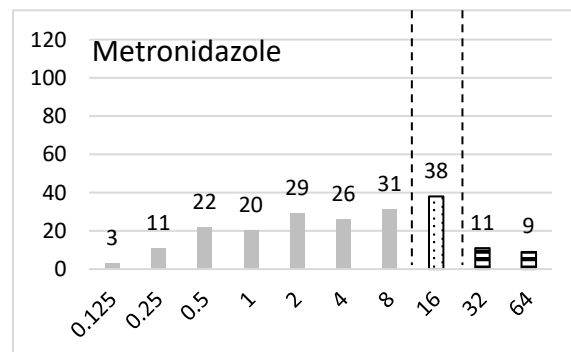
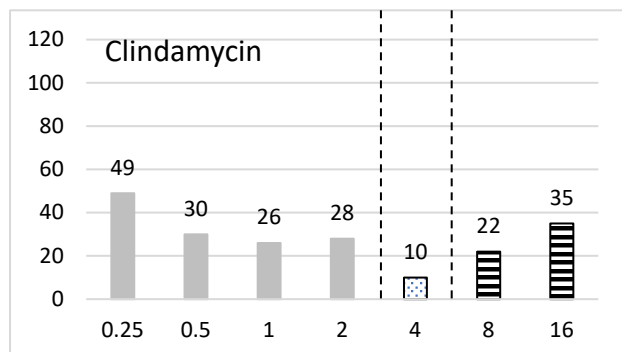
**Table S3 (A):** PCR reagents and reaction setup for *nim* gene and *cfiA* gene

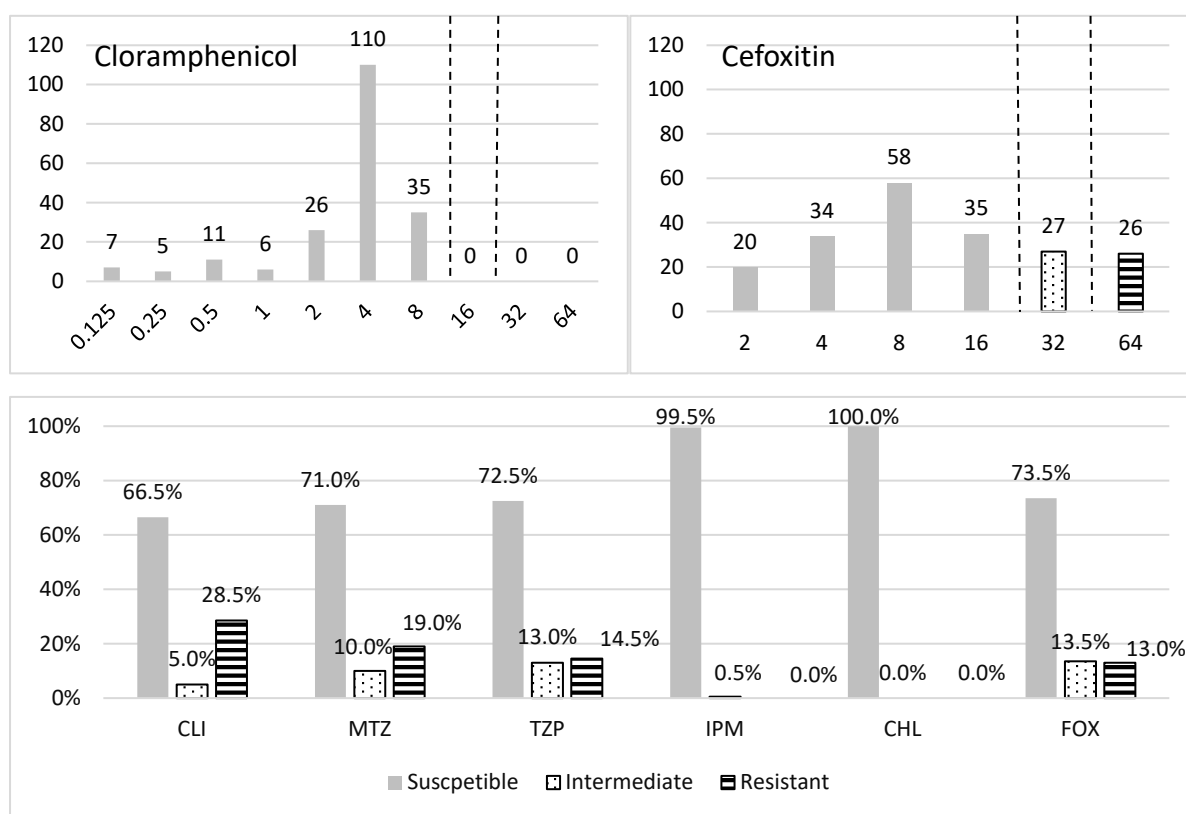
Reagents	Volume for 25 µl
PCR buffer	0.5 µl
dNTP	1.25 µl
Forward primer	0.5 µl
Reverse primer	0.5 µl
Taq polymerase	0.25 µl
Template DNA	02 µl

MgCl <sub>2</sub>	01 µl
dH <sub>2</sub> O	19 µl
Total	25 µl

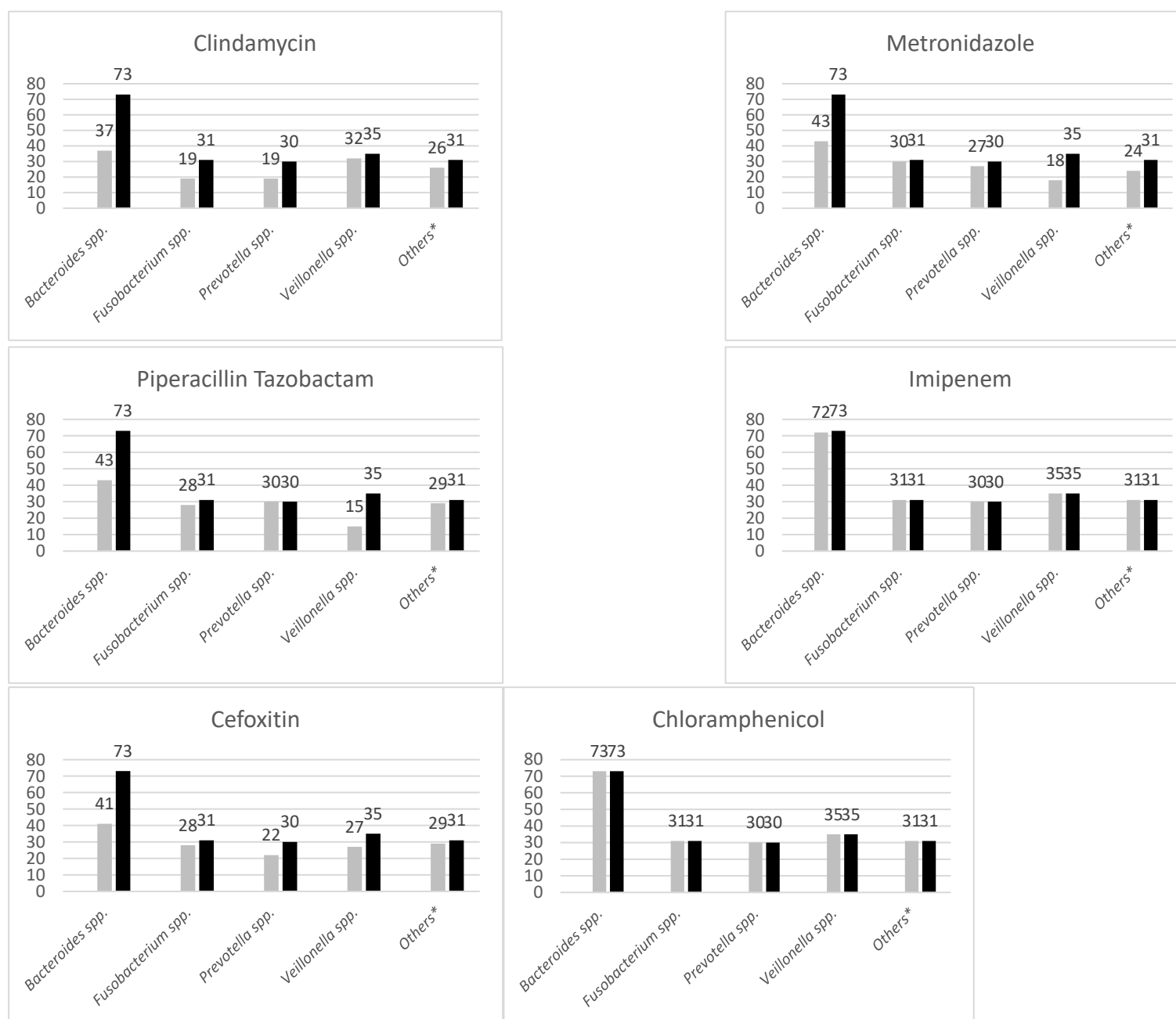
**Table S3 (B):** PCR reagents and reaction setup with master mix for genes (*cfiA*, *ermF*, *cfxA*, *cepA*, *cat*), IS1186 and *cfiA*<sup>IS</sup>

Reagents	Volume for 20 µl
Master mix (Sigma REDTaq ReadyMix™ PCR Reaction Mix)	10 µl
Forward primer	0.4 µl
Reverse primer	0.4 µl
Template DNA	02 µl
dH <sub>2</sub> O	7.2 µl
Total	20 µl

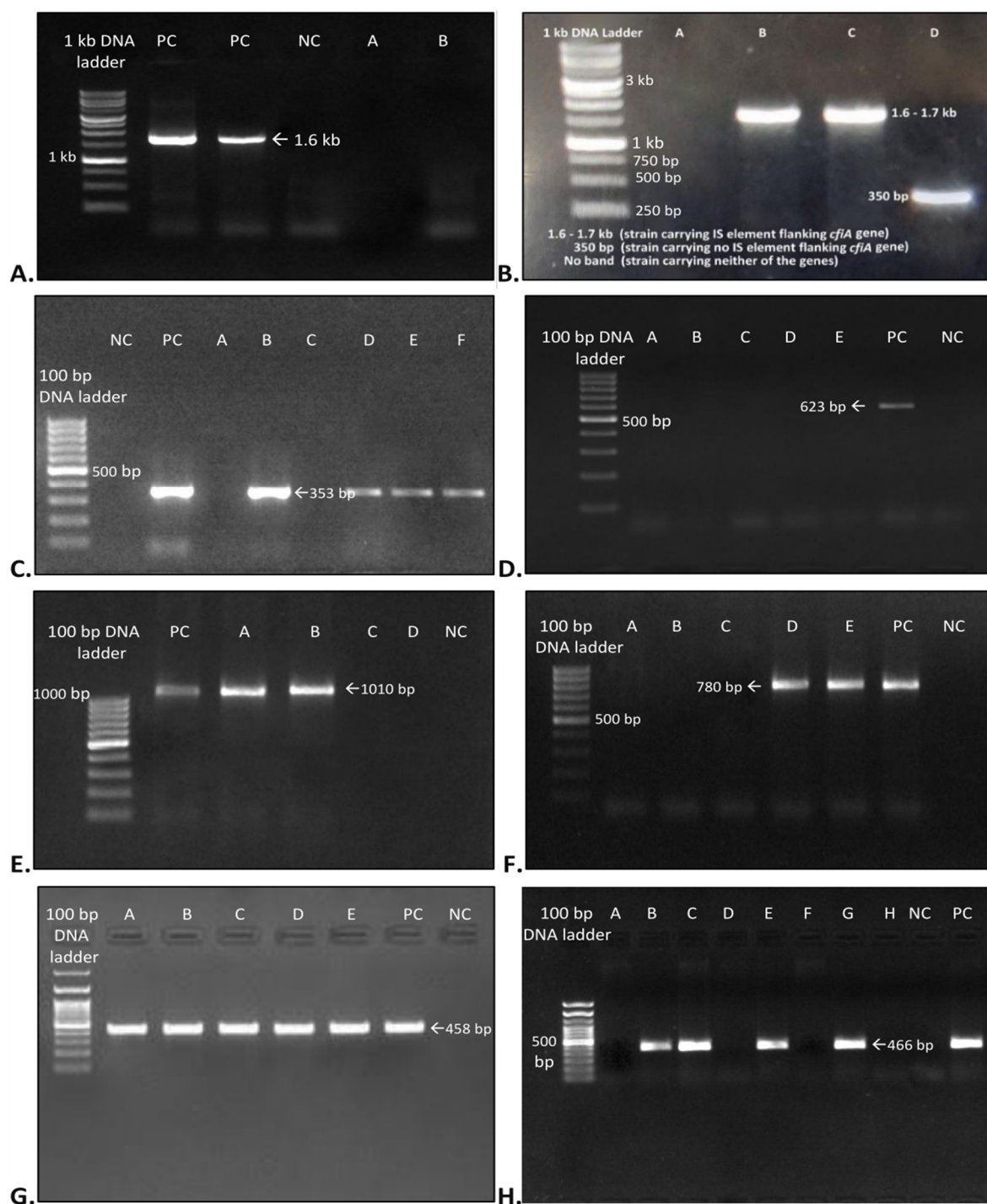




**Figure S1:** MIC distribution of anaerobic isolates and the overall resistance rate to six antimicrobials. Bars depict the numbers of resistant, intermediate, and susceptible isolates to clindamycin (CLI), metronidazole (MTZ), piperacillin/tazobactam (TZP), imipenem (IPM), chloramphenicol (CHL), cefoxitin (FOX) at different drug concentrations. The broken lines represent clinical breakpoints (mg/L) as per the CLSI guidelines. The x-axis shows drug concentration in (mg/L), and the y-axis shows the number of isolates.



**Figure S2:** Antimicrobial susceptibility of clinical isolates representing different genera to tested antimicrobials. Others\* *Acidaminococcus* spp., *Alistipes* spp., *Anaerobiospirillum* spp., *Bilophila* spp., *Parabacteroides* spp., *Porphyromonas* spp., *Sutterella* spp. The black and grey bars represent total number of isolates and susceptible isolates, respectively.



**Figure S3:** Agarose gel picture for PCR amplified product of A.) *IS1186* element B.) *cfiA* gene and the flanking IS elements C.) *cfiA* gene D.) *cat* gene E.) *cfxA* gene F.) *cepA* gene G.) *nim* gene H.) *ermF* gene. The first lane contains DNA ladder; PC, positive control; NC, negative control.