

**Supplementary Table 2.** The list and sequences of CARD-FISH probes, competitors and helpers (H) used to verify the ARNIS approach. FA% – concentration of formamid in the hybridization buffer (at 37°C).

Probe	Target Group	Probe Sequence (5'-3')	Competitor/Helper Sequence (5'-3')	FA%	Reference
HGC69a	Actinobacteria	TATAGTTACCACCGCCGT	none	30	Roller et al 1994
Ac1-852	acI lineage of Actinobacteria	AATGCGTTAGCTGCGTCGCA	H1: AAACCGTGGAAGGTYCSCACA H2: TCCCCAGGCGGGGCRCTT	55	Warnecke et al 2005
ALF968	Alphaproteobacteria	GGTAAGGTTCTGCGCGTT	none	55	Neef 1997
CF968	Bacteroidetes	GGTAAGGTTCCCTCGCGTA	none	55	Acinas et al 2014
BET42a	Betaproteobacteria	GCCTTCCCACCTTCGTTT	Competitor: GCCTTCCCACATCGTTT	55	Manz et al 1992
CF319a	Cytophaga- Flavobacteria	TGGTCCGTGTCTCAGTAC	none	55	Manz et al 1996
R-BT065	<i>Limnohabitans</i> spp.	GTTGCCCCCTCTACCGTT	none	55	Šimek et al 2001

## References

- Acinas SG, Ferrera I, Sarmiento H, Diez-Vives C, Forn I, Ruiz-Gonzalez C et al (2015). Validation of a new catalysed reporter deposition-fluorescence in situ hybridization probe for the accurate quantification of marine Bacteroidetes populations. *Environ Microbiol* 17: 3557-3569.
- Manz W, Amann R, Ludwig W, Wagner M, Schleifer KH (1992). Phylogenetic Oligodeoxynucleotide Probes for the Major Subclasses of Proteobacteria - Problems and Solutions. *Syst Appl Microbiol* 15: 593-600.
- Manz W, Amann R, Ludwig W, Vancanneyt M, Schleifer KH (1996). Application of a suite of 16S rRNA-specific oligonucleotide probes designed to investigate bacteria of the phylum cytophaga-flavobacter-bacteroides in the natural environment. *Microbiology* 142: 1097-1106.
- Neef A (1997). Anwendung der in situ Einzelzell-Identifizierung von Bakterien zur Populationsanalyse in komplexen mikrobiellen Biozönosen, Technische Universität München.
- Roller C, Wagner M, Amann R, Ludwig W, Schleifer KH (1994). In situ probing of Gram-positive bacteria with high DNA G+C content using 23S rRNA-targeted oligonucleotides. *Microbiology* 140: 2849-2858.
- Šimek K, Pernthaler J, Weinbauer MG, Horňák K, Dolan JR, Nedoma J et al (2001). Changes in bacterial community composition and dynamics and viral mortality rates associated with enhanced flagellate grazing in a mesoeutrophic reservoir. *Appl Environ Microbiol* 67: 2723-2733.
- Warnecke F, Sommaruga R, Sekar R, Hofer JS, Pernthaler J (2005). Abundances, identity, and growth state of actinobacteria in mountain lakes of different UV transparency. *Appl Environ Microbiol* 71: 5551-5559.