



Article

It takes two to tango: a bacterial biofilm provides protection against a fungus feeding bacterial predator

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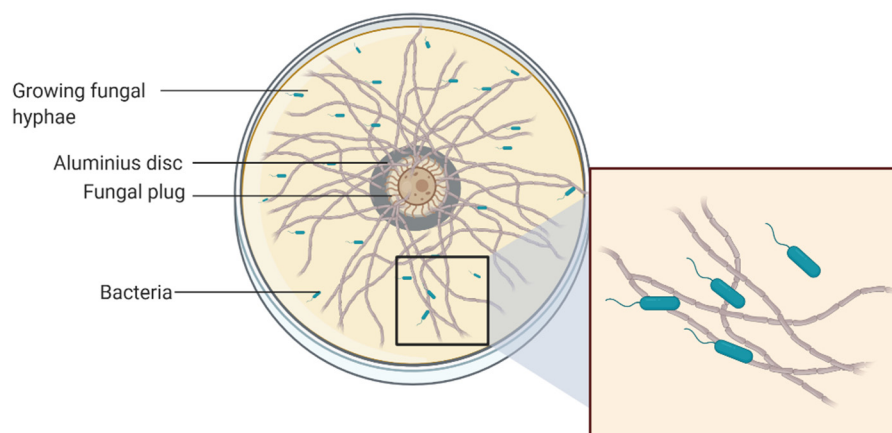


Figure S1: *In vitro* mycophagy assay set up. *Collimonas fungivorans* or *Kosakonia radicincitans* were grown on a Petri dish containing phytigel medium. A fungal plug from an actively growing *Serendipita indica* culture was placed in the center of the Petri dish on an aluminum disc. While expanding radially, growing fungal hyphae were confronted with bacteria.

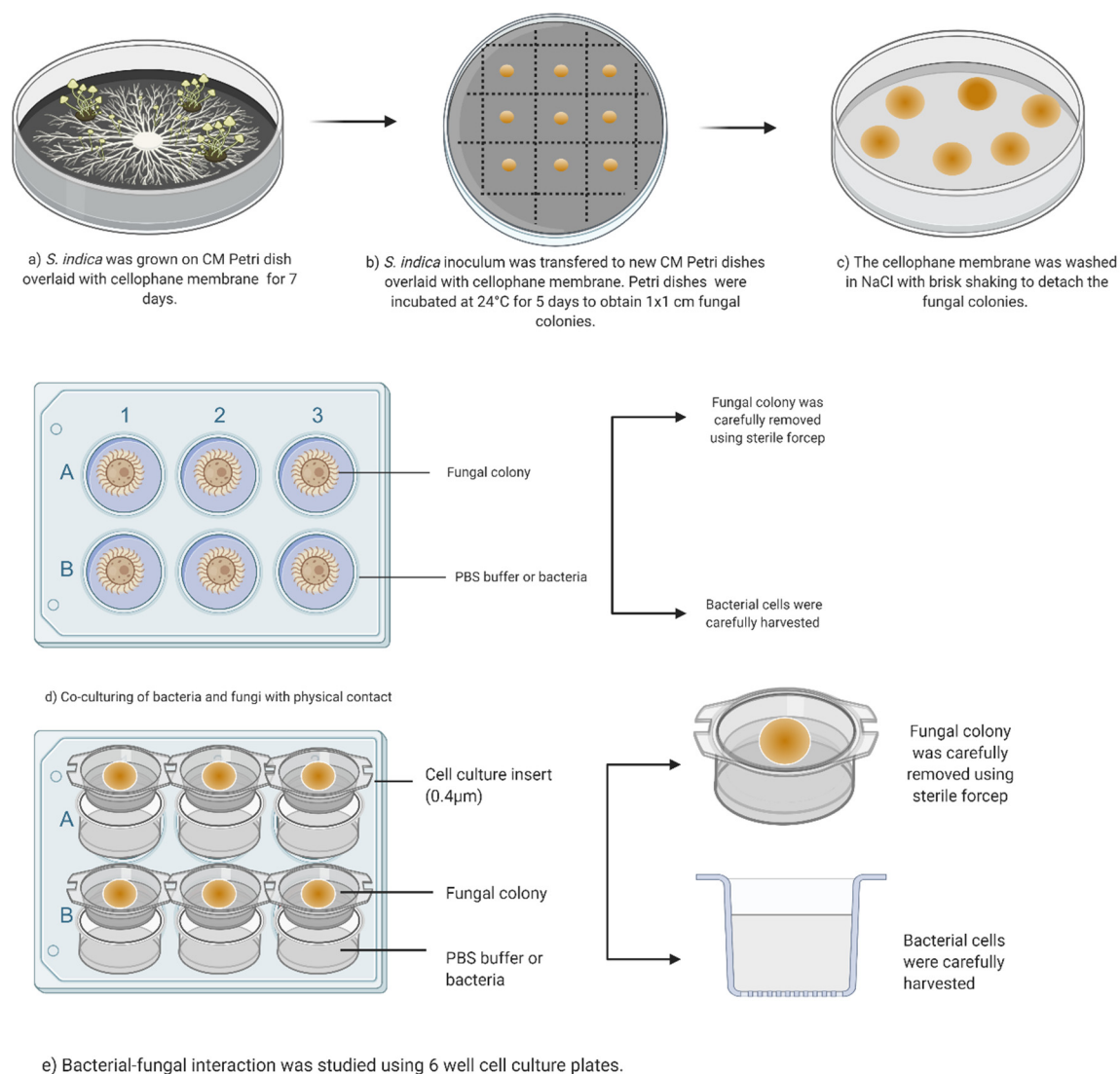


Figure S2: Schematic diagram showing (a-c) culturing of *Serendipita indica* fungal colonies on Petri dishes containing solidified CM overlaid with cellophane membrane and detachment of fungal colonies from the cellophane membrane or (d-e) co-culturing of fungus with *Collimonas fungivorans* or *Kosakonia radicincitans* using 6-well plates with or without cell culture inserts.

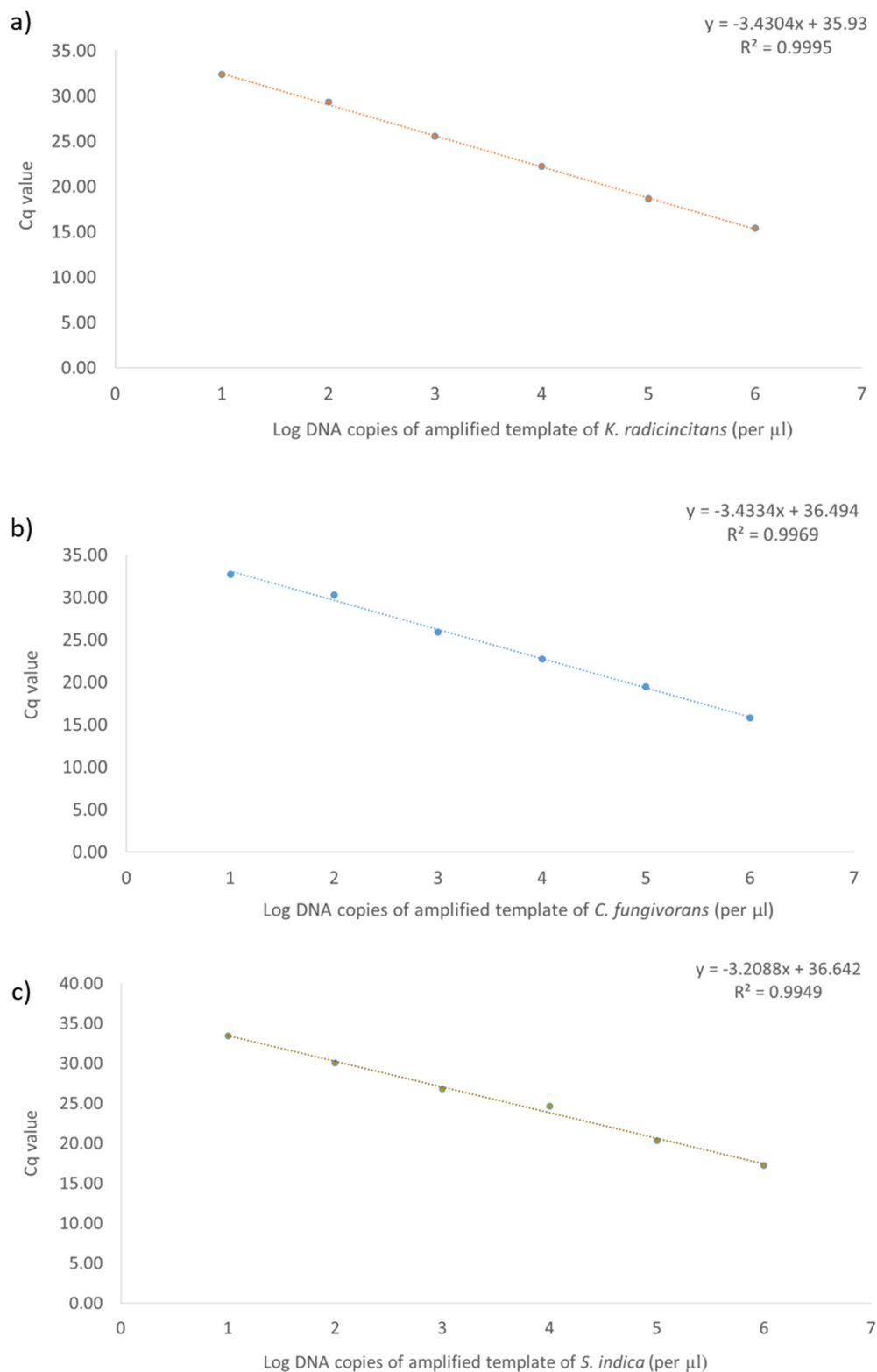


Figure S3: qPCR-generated standard curves of a) *Kosakonia radicincitans* and b) *Collimonas fungivorans* for absolute quantification by TaqMan assay using *Kosakonia* and *Collimonas* specific dual-labelled probes. c) Standard curve of *Serendipita indica*, which was quantified by SYBR green I assay.

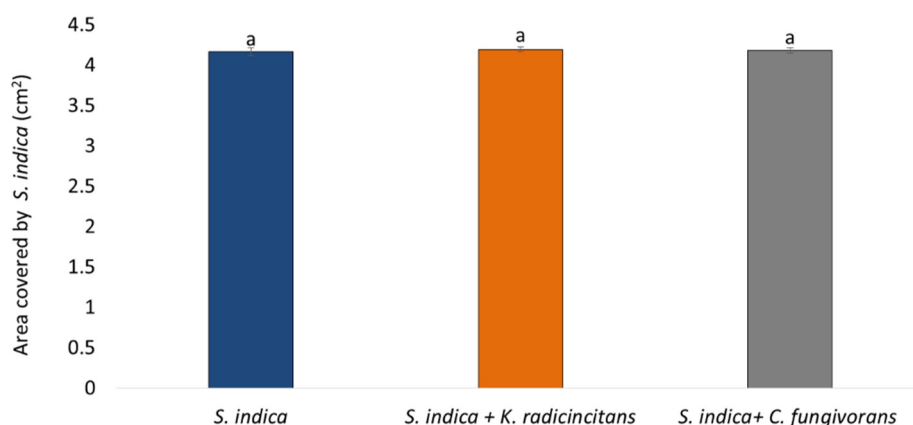


Figure S4: Area of *Serendipita indica* colonies (cm²) after three weeks of growth. *S. indica* was confronted with either *Kosakonia radicincitans* or *Collimonas fungivorans* on water yeast agar plates. Same letters above bars represent no significant differences ($P \leq 0.05$) according to one way ANOVA followed by a Tukey's range test. Error bars indicate standard deviation of means of three different replicates.

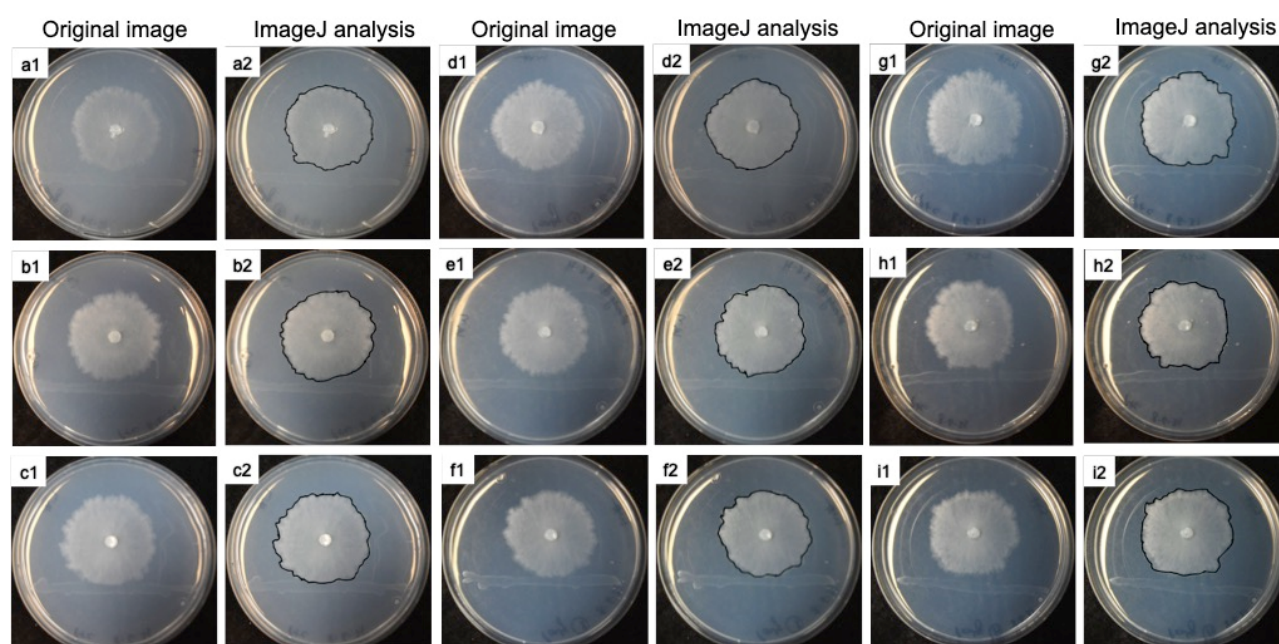


Figure S5: Petri dishes showing the area covered by uninoculated *S. indica* when confronted with the fungus-feeding bacterium *C. fungivorans*. (a1-i1) Original image of the *S. indica* colony growing in the Petri dishes. (a2-i2) Images analyzed by ImageJ software, used to measure the area covered by *S. indica* in the Petri dishes.

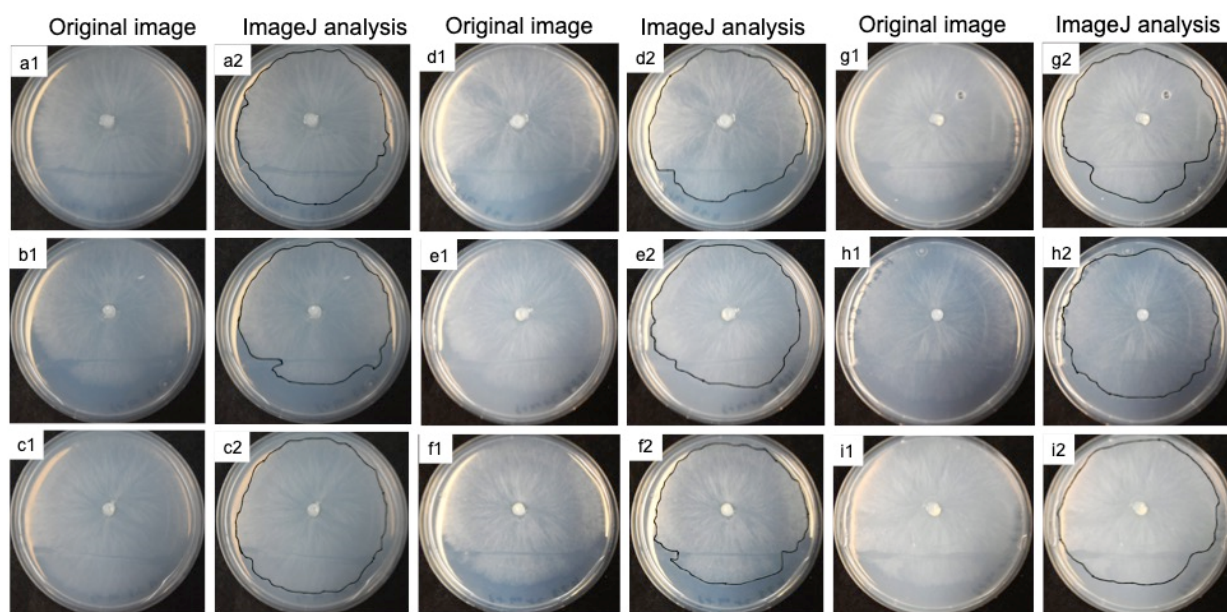


Figure S6: The area covered by *S. indica*-*K. radicincitans* consortia when confronted with the fungus-feeding bacterium *C. fungivorans*. (a1-i1) Original image of the *S. indica* colony growing on the Petri dishes. (a2-i2) Images analyzed by ImageJ software, used to measure the area covered by *S. indica* on the Petri dishes.

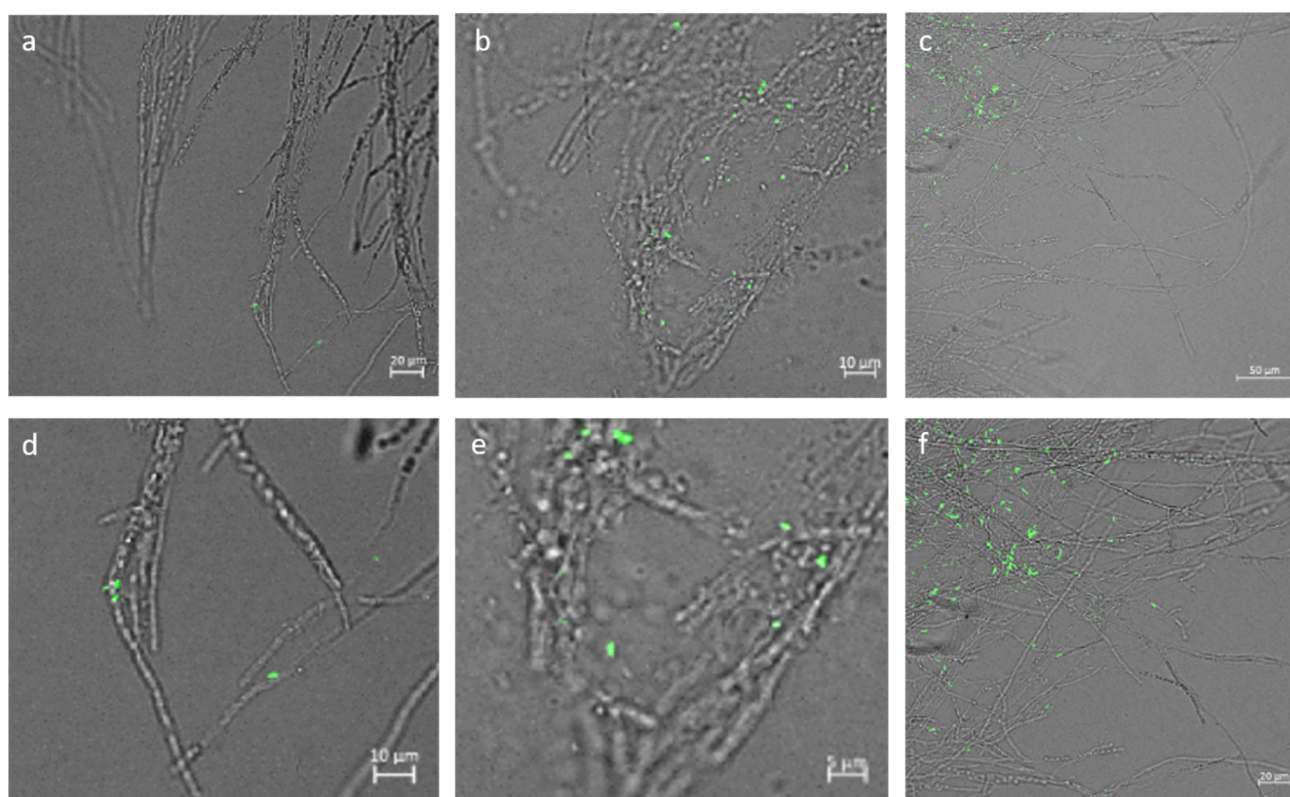


Figure S7: Imaging of microcolonies of *Serendipita indica* co-cultured with *Kosakonia radicincitans* for 72h at 24°C. The colonization of the fungal hyphae by *K. radicincitans* was observed microscopically. (a-c) *K. radicincitans* colonized the hyphae of *S. indica* forming bacterial aggregates 4h post inoculation. (d-e) *K.*

radicincitans aggregates found attached to the hyphal tips of *S. indica*. The GFP strain of *K. radicincitans* was used and *S. indica* is shown in grey scale. Pictures were taken using a confocal microscope at 40x magnification.

Table S1. List of primers and probe sequences used in this study

Primer/Probes	Sequence (5'-3')	Reference
Tef-f	ACCGTCTTGGGGTTGTATCC	Bütehorn et al., 2000
Tef-r	TCGTCGCTGTCAACAAGATG	Bütehorn et al., 2000
Eddy3for	GTACAGAATCCCGAAGAGATTTGG	Ogawa-Höppener et al., 2007
Eddy3rev	ACTTAACCCAACATCTCACGACA	Ogawa-Höppener et al., 2007
TaqMan probe Sophie	FAM-CGAAAGAAAGCTGTAACACAGG-BHQ	Ogawa-Höppener et al., 2007
519f	CAGCMGCCGCGGTAANWC	Lane et al., 1991
E.radr	CGTGGACTACCAGGGTATCTAATC	Ruppel et al., 2006
E.rad TaqMan probe	[6-FAM] CTCCCCACGCTTTCGCACCTGAGC [BHQ-6-FAM]	Ruppel et al., 2006