

SUPPLEMENTARY INFORMATION

Bacterial-specific induction of inflammatory cytokines significantly decreases upon dual species infections of implant materials with periodontal pathogens in a mouse model

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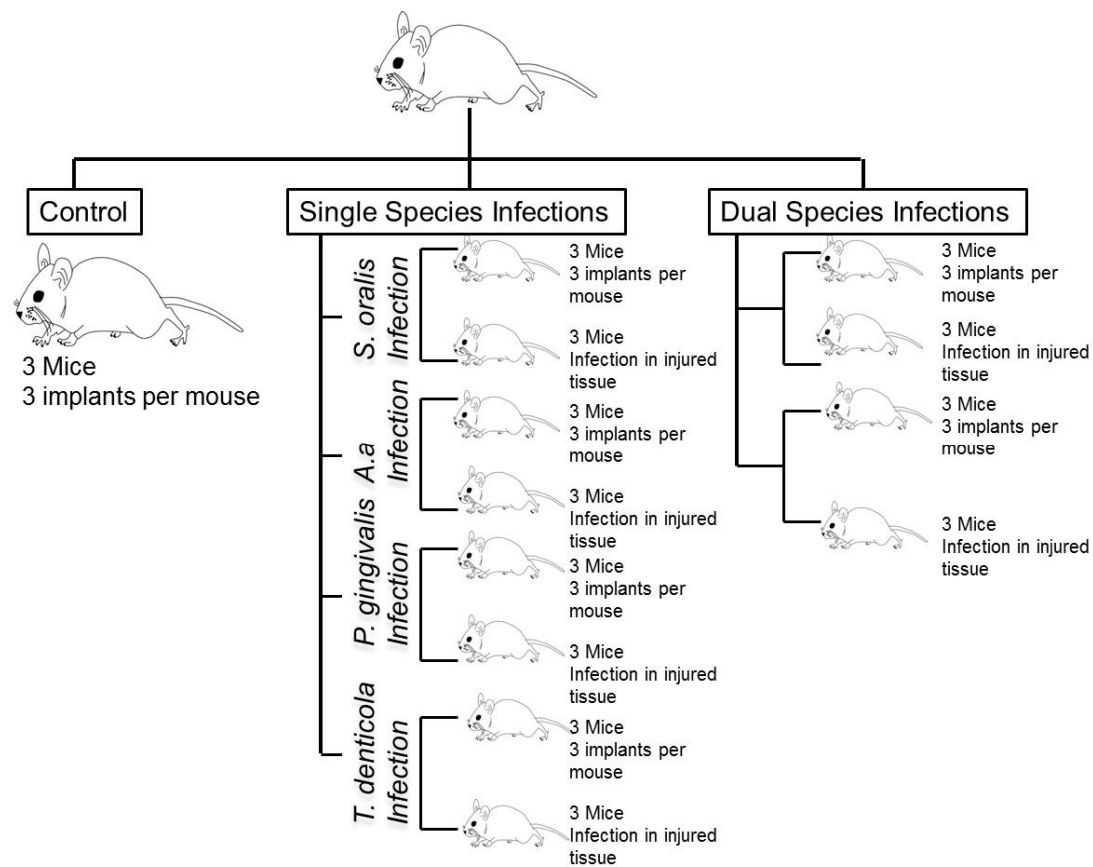


Figure S1. Classification of mice into respective groups for the measurement of cytokines.

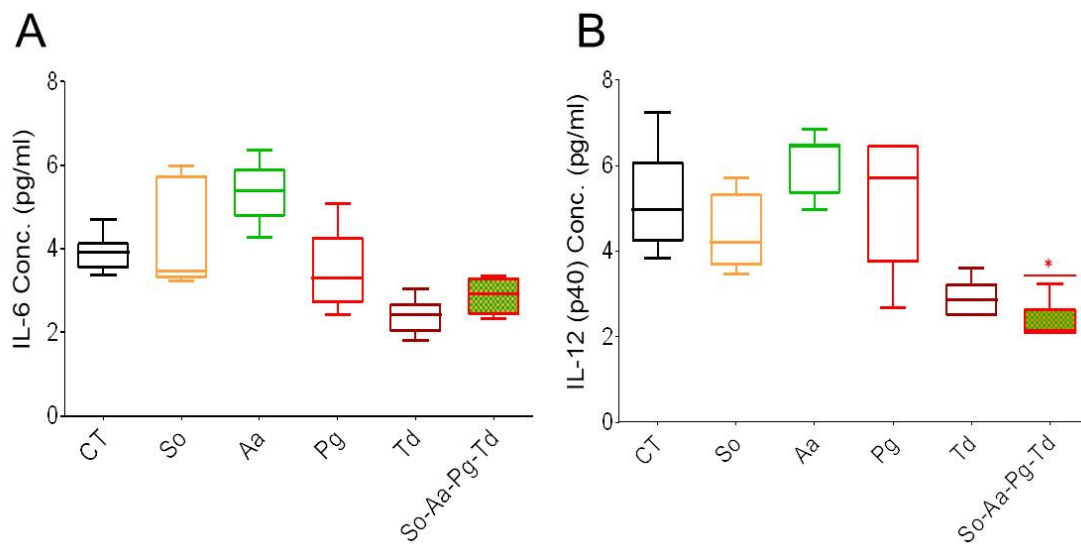


Figure S2. Expression of interleukins IL-6 (A) and IL-12 (B) in murine fibroblast.

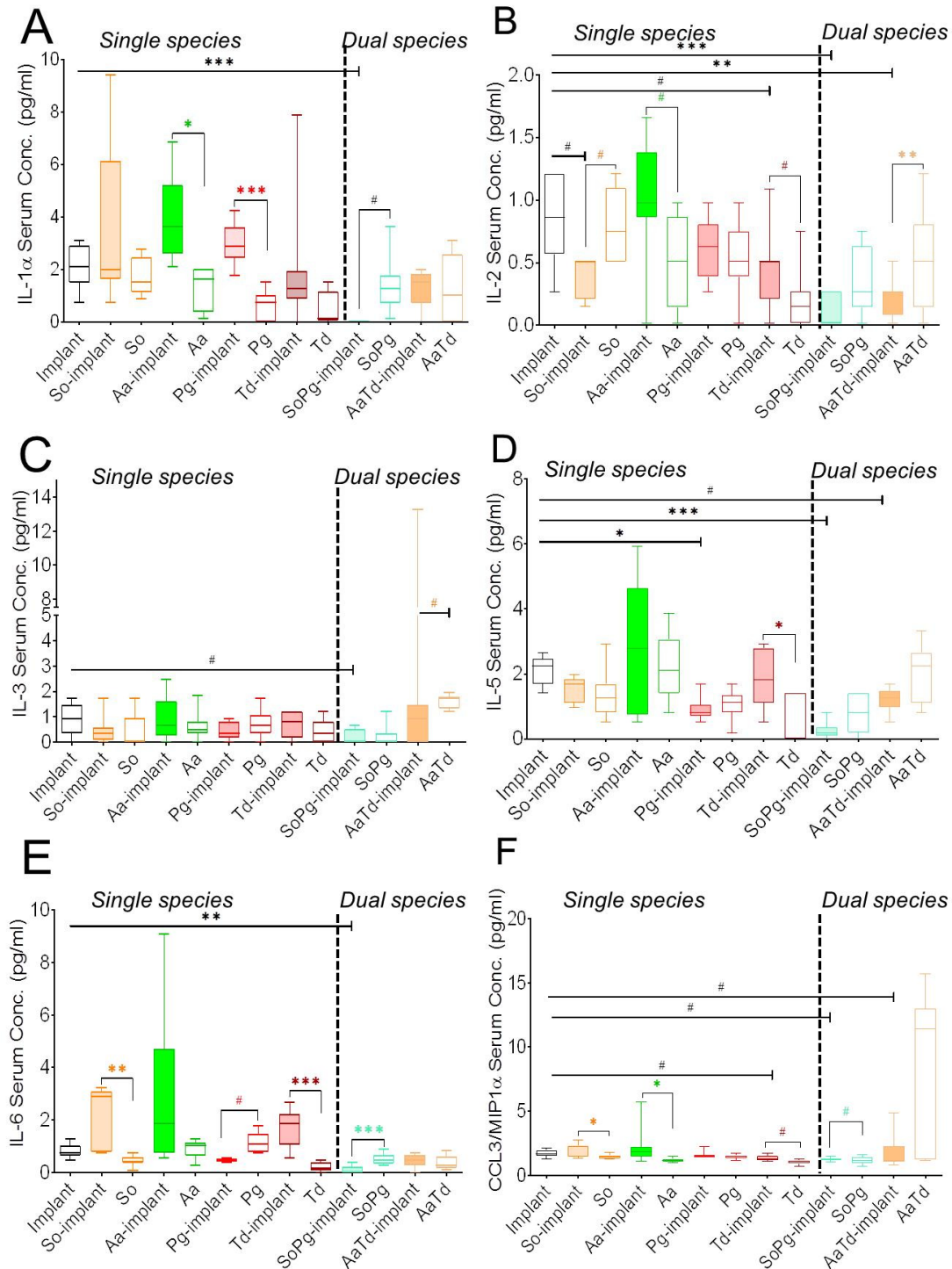


Figure S3. Systemic profiles of inflammatory cytokines 21 days post-implantation. Systemic cytokine expressed in the concentration of 0-10 pg/mL for IL-1 α (A), IL-2 (B), IL-3 (C), IL-5 (D), IL-6 (E), CCL3/MIP1 α (F) were determined in the blood serum of mice carrying infected implants.

Supplementary Table S1. List of cytokines in pictograms per microliter (pg/ml) expressed near or below detection limits. (biological replicates (n=3), technical replicates (3) each)

	IL-1b	IL-4	IL-10	GM-CSF
Sterile implant	0.57	0.27	2.92	-
	0.57	0.27	0.83	
	0.57	0.09	2.92	
	1.99		0.83	
implant + <i>S. oralis</i>	0.57	0.45	0.83	-
	0.57		2.92	
	3.15			
	0.57			
	1.99			
<i>S. oralis</i> infection	0.57	0.45	2.92	-
	0.57			
	0.57			
	0.57			
Implant + <i>A. actinomycetemcomitans</i>	1.99	0.62	6.51	-
	3.15	1.31	0.83	
	2.59	2.99	3.86	
		0.45	4.77	
			0.83	
<i>A. actinomycetemcomitans</i> infection	0.57	-	0.83	-
	0.57			
Pg-implant	0.57	0.27	6.51	-
	1.99	0.27	6.51	
			0.83	
			2.92	
			4.77	
			1.92	
Pg	0.57	-	0.83	-
			0.83	
Td-implant	0.57	1.14	-	-
Td	-	-	4.77	-
SoPg-implant	-	-	0.83	-
			0.83	
			2.92	
SoPg	-	-	0.83	-

AaTd-implant	0.57	1.48	0.83	7.76
AaTd	-	1.90	0.83	10.68
		1.48	2.92	1.13
		1.65	6.51	7.76
		1.31	2.92	14.58
			8.17	9.77
			2.92	

Supplementary Table S2. Dual vs single species infections: Cytokine expression decreases with dual species infections compared to single species infections

Cytokines	SoPg vs So	SoPg vs Pg	SoPg-implant vs So-implant	SoPg-implant vs Pg-implant	AaTd- vs Aa	AaTd- vs Td	AaTd-implant- vs Aa-implant	AaTd-implant- vs Td-implant
IL-1 α			↓***	↓***			↓*	
IL-2	↓#			↓*	↑#			↓***
IL-3		↓#			↑#	↑**		
IL-5				↓*		↑**		
IL-6		↓#	↓***	↓#		↓#	↓*	↓*
IL-9		↓***	↓#	↓#		↑*		
IL-12(p40)	↑#	↑***	↓**	↓***			↓#	↓*
IL-12(p70)		↓#			*		↑	
IL-13			↓#	↓#		↓#	↓#	↓#
CXCL1/KC			↓***		↓*		↓#	
CCL5/RANTES	↑#	↑*	↓#			↑**	↓#	↓#
CCL-3/MIP1 α		↓#	↓#	↓#	↑**	↑***	↓#	
CCL11			↓#					
IFN- γ		↓***	↓#	↓#		↑**		
G-CSF				↓*	↓#		↓***	↓**
TNF- α		↓#				↑*		
CCL2/MCP-1	↓*		↓***	↓#			↓#	↓#
Upregulated	2	2	0	0	3	8	0	0
Downregulated	2	7	11	11	2	2	9	7

Statistically significant increase (↑) or decrease (↓) in the serum cytokine levels at ***, **, *, and # indicated $p < 0.001$, $p < 0.01$, $p < 0.05$, and significant p value prior to Bonferroni correction, respectively, between implants and infected implants in mice upon single or dual species infections.