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Supplementary Information for

Streptococcus gordonii programs epithelial cells to resist ZEB2 induction by *Porphyromonas gingivalis*

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Table S1

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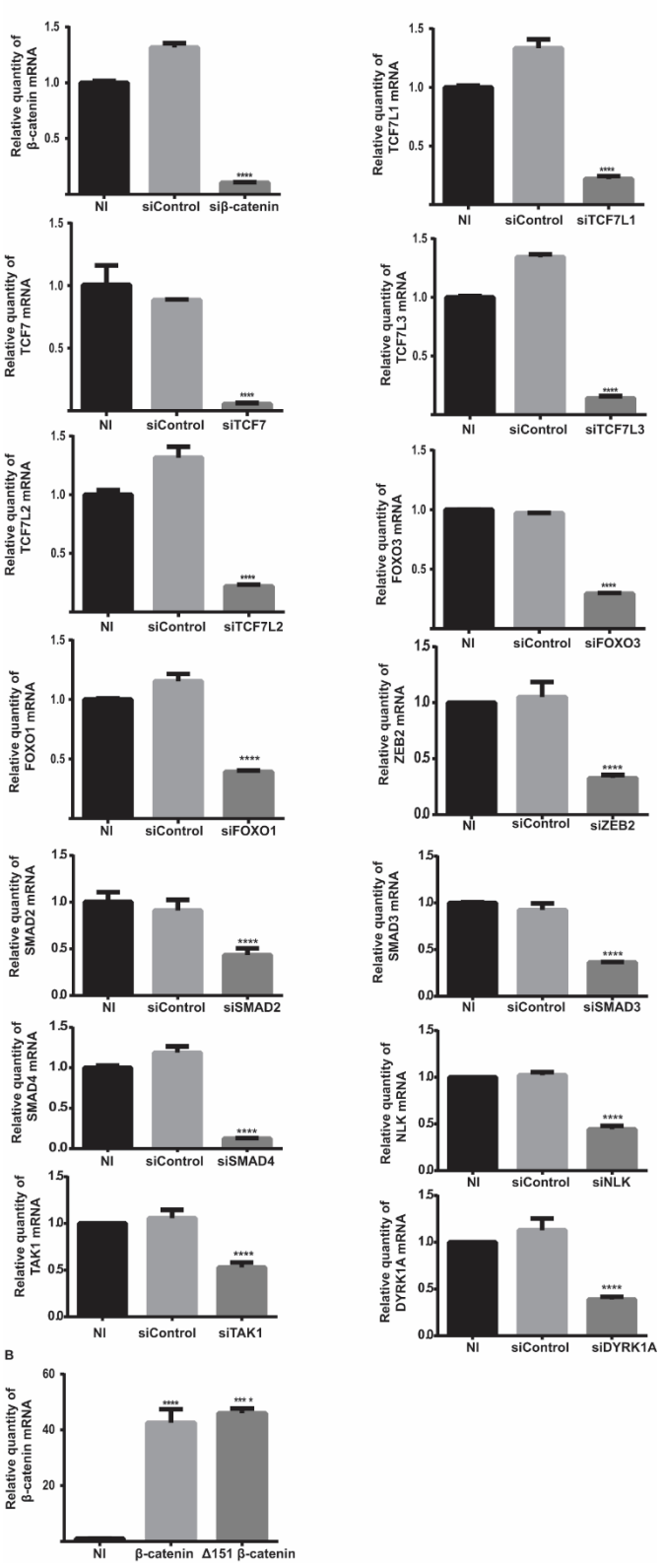

Fig. S1. Confirmation of siRNA knockdowns and knockins by qRT-PCR. **** $P < 0.001$.

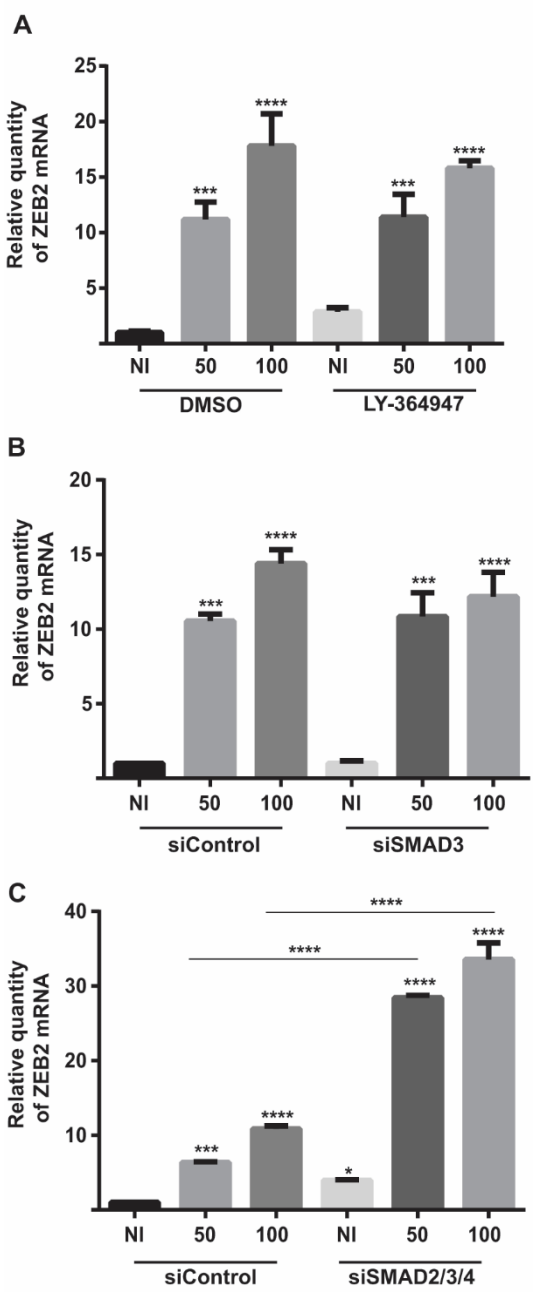

Fig. S2. ZEB2 regulation by *P. gingivalis* does not involve the TGF-β pathway. TIGKs were challenged with *P. gingivalis* 333277 for 24 h at the MOI indicated. ZEB2 mRNA levels were measured by qRT-PCR. Data were normalized to GAPDH mRNA and are expressed relative to noninfected (NI) controls. (A) TIGK cells were incubated with LY-364947 (20 μM), or vehicle control (DMSO) for 2 h prior to *P. gingivalis* challenge. (B and C) TIGK cells were transiently transfected with siRNA to SMAD3 (B) or SMAD2/3/4 (C) prior to *P. gingivalis* challenge. Controls were scrambled siRNA (siRNA control). Data represent three independent experiments with three replicates. Error bars represent the SEM. * $P > 0.05$, *** $P < 0.005$, **** $P < 0.001$ compared to NI unless indicated.

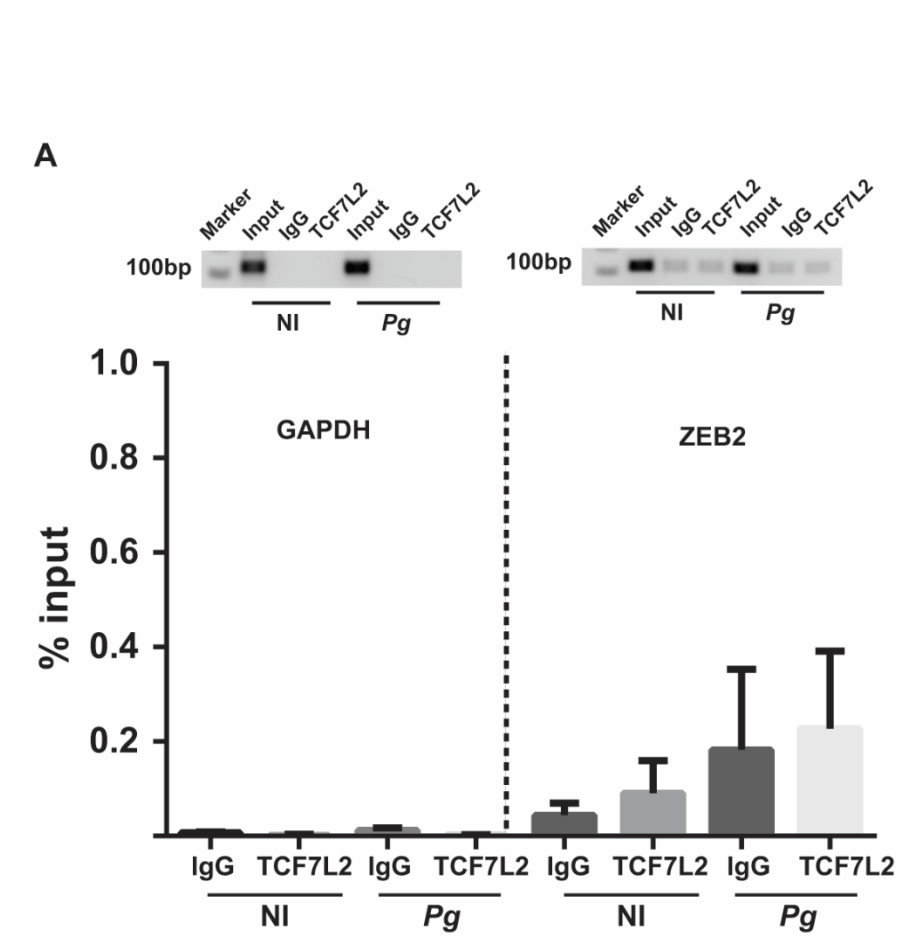

Fig. S3. TIGK cells were challenged with *P. gingivalis* MOI:100 for 24 h, or left uninfected (NI), and subjected to chromatin immunoprecipitation (ChIP) using anti-TLF7L2 IgG. The precipitated DNA was subsequently analyzed by end point PCR and by qPCR with primers to the ZEB2 promoter region or the GAPDH promoter as a control. qPCR was expressed relative to the input DNA. Data represent three independent experiments with three replicates. Error bars represent the SEM.

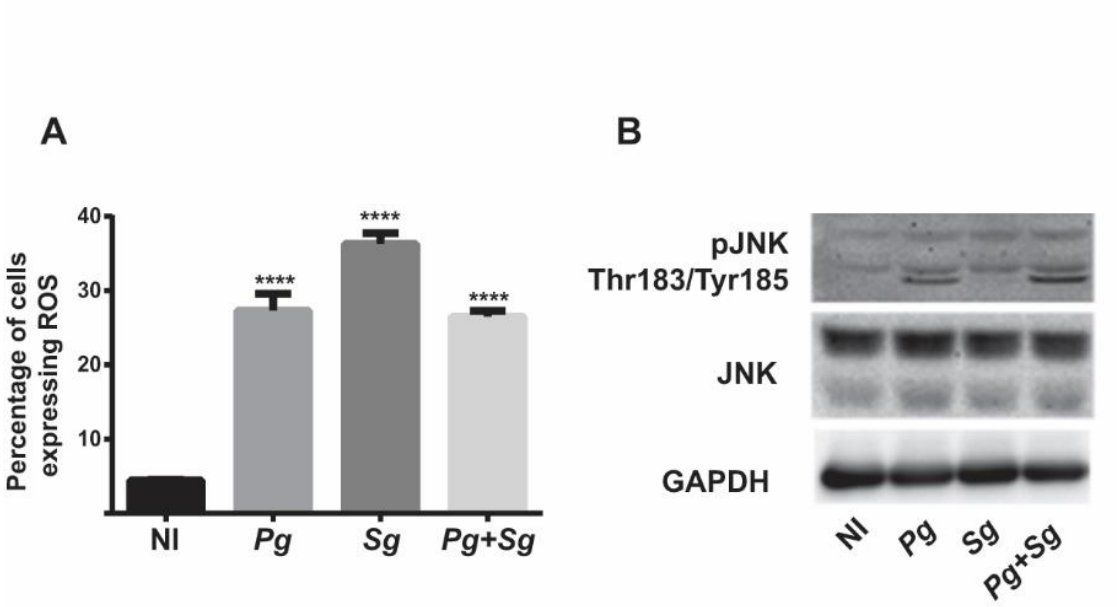

Fig. S4. *S. gordonii* does not impede *P. gingivalis*-induced increases in ROS or activation of JNK. TIGK cells were challenged with *P. gingivalis* (Pg) and/or *S. gordonii* (Sg) at MOI:10 for each strain for 30 min. Control cells were non-infected (NI). (A) TIGKs were stained with APF, and ROS levels were determined by flow cytometry. (B) Immunoblot of lysates of TIGK cells probed with the antibodies indicated. Control cells were NI. GAPDH was used as a loading control. Results are representative of three independent experiments. **** $P < 0.001$ compared to NI.

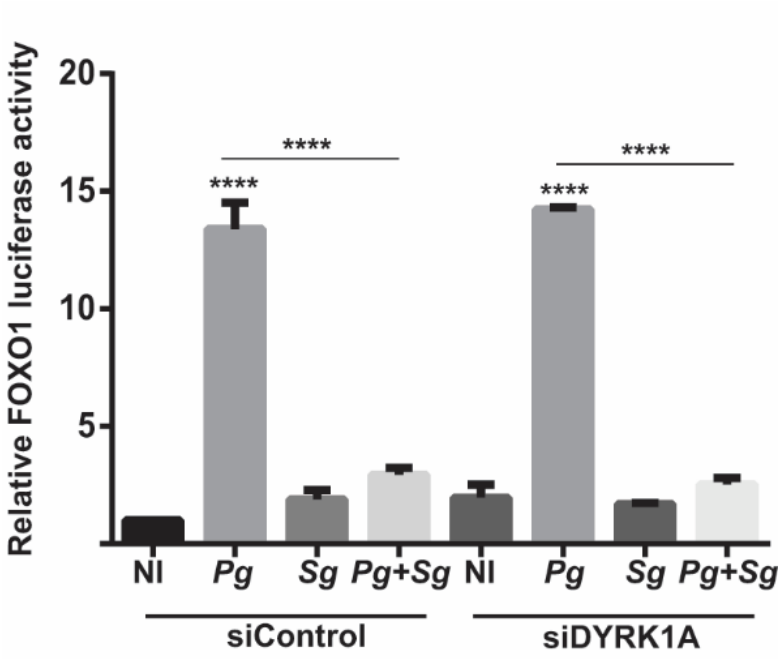

Fig. S5. Knockdown of DYRK1A does not impede *S. gordonii* antagonism of FOXO1 activation by *P. gingivalis*. TIGK cells were transiently transfected dually siDYRK1A, along with the FOXO promoter–luciferase reporter plasmid, or a constitutively expressing Renilla luciferase reporter. FOXO luciferase activity after 15 min of bacterial challenge was normalized to the level of Renilla luciferase. Data represent three independent experiments with three replicates. Error bars represent the SEM. **** $P < 0.001$ compared to NI unless indicated.

Table S1. Primers used in this study

Primer name	Sequence	Resulting plasmids and descriptions
hCTNNB1-qPCR-F	5'- CTTACACCCACCATCCCACT -3'	Quantitative RT-PCR
hCTNNB1-qPCR-R	5'- TGCACGAACAAGCAACTGAA -3'	
hTCF7-qPCR-F	5'- TACTCCGCCTTCAATCTGCT -3'	
hTCF7-qPCR-R	5'- CTTGCTTCTGGCTGATGTCC -3'	
hTCF7L1-qPCR-F	5'- TGAAGGAGATGAGGGCCAAG -3'	
hTCF7L1-qPCR-R	5'- CTTCTCGAGACAGGTTGTGC -3'	
hTCF7L2-qPCR-F	5'- CAGGAATCGTCCCAGAGTGA -3'	
hTCF7L2-qPCR-R	5'- CACTCAGCTACGACCTTTGC -3'	
hGAPDH-qPCR-F	5'- GCTCAGACACCATGGGGAAG -3'	
hGAPDH-qPCR-R	5'- GAACATGTAAACCATGTAGTTGAGG -3'	
hCTNNB1-Exp-F	5'- GGATCCACCATGGCTACTCAAGCTGATTTG -3'	β-catenin expression vector
hDN151-CTNNB1-Exp-F	5'- GGATCCACCATGGCAATCCCTGAACTGACAA -3'	
hCTNNB1-Exp-R	5'- GCGGCCGCTTACAGGTCAGTATCAAACCAG -3'	
hTCF7L1-Exp-F	5'- GAATTCACCATGCCCCAGCTCGGCG -3'	TCF7L1 expression vector
hTCF7L1-Exp-R	5'- GCGGCCGCTTAGTGGGCAGACTTGGTGACCAGG -3'	
ZEB2-pro-1366-F	5'- GAGCTCCATCAGACTGGAAACAGGAGGGTG -3'	ZEB2 reporter plasmid
ZEB2-pro-1054-F	5'- GAGCTCTAGTTCCAATTATGTGCAGTG -3'	
ZEB2-pro-963-F	5'- GAGCTCCTGGGAAGTGGGCTCTGAATTGG -3'	
ZEB2-pro-551-F	5'- GAGCTCGAGGTGTAGAGAGATTCAGAGATCGG -3'	
ZEB2-pro-316-F	5'- GAGCTCCCAGAAGCTGTACTGAGATACCTACAC -3'	
ZEB2-pro-3041-R	5'- GTCGACTGATAAGAGCGGATCAGATGGCAGTTCG -3’	
ZEB2-pro-Mut1-F	5'- AAGAAAAGGGTAACAATAAGAGAAAGGGCA -3'	
ZEB2-pro-Mut1-R	5'- TTTAGGTACCAGAGCCAGA -3'	
ZEB2-pro-Mut2-F	5'- TTGGCTGTCTTTAGGGACTTCTATATTAGT -3'	
ZEB2-pro-Mut2-R	5'- ATTTGAGGATGCTGAGCACA -3'	
ZEB2-Chip-F	5'- GGGCAGAGAACTTTGTTCCA -3’	ChIP assay
ZEB2-Chip-R	5’ –CCGGGTCCTCAATCCAGATA -3’	
GAPDH-Chip-F	5'- CCACATCGCTCAGACACCAT -3’	
GAPDH-Chip-R	5'- CCCGCAAGGCTCGTAGAC -3’	
FOXO1-E-F	5’- CATCCTGTTCGGTCATAATGGGCTCGAGTCTCCCACTAATAGTACTA -3’	FOXO1 S329E mutation
FOXO1-E-R	5’- TAGTTA5’TTAGTGGGAGACTCGAGCCCATTATGACCGAACAGGATG -3’	
FOXO1-A-F	5’- TTCGGTCATAATGGGTGCGAGTCTCCCACTAATAG -3’	FOXO1 S329A mutation
FOXO1-A-R	5’ –CTATTAGTGGGAGACTCGCACCCATTATGACCGAA -3’	