

1 **Supplementary Materials**

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3 **Supplementary Figure Legends**

4 **Figure S1. CARD9 expression in the colon tissue. Related to Figure 1. (A)** RT-PCR
5 analysis for the expression of *Card9* and *I18* in the CD45⁺ or EpCAM⁺ cells purified from the
6 colons of WT mice at indicated days after AOM injection by FACS. **(B)** Gating strategy for the
7 EpCAM⁺ and CD45⁺ cells in colon. **(C)** Immunofluorescence for CARD9, EpCAM and CD11b in
8 the colon tissues at indicated days after AOM injection. Scale bar = 20um. Data represent two
9 independent experiments with 6-10 mice per group per time point. **(D)** Immunohistochemistry for
10 CARD9 in human colon tissue biopsy from protein atlas. **(E)** Expression of ITGAM (CD11b) in
11 the normal and tumor colon biopsies in the GENT database. **(F)** Schematic representation of
12 AOM-DSS treatment. Mice were injected with AOM on day 0, and given 2% DSS in drinking
13 water on day 5 for 6 days, followed by drinking water. Mice were given two subsequent rounds
14 of DSS with an interval of 17 days. Error bars represent mean±standard error of the mean.

15 **Figure S2. CARD9 does not affect goblet cell differentiation or mucus production early**
16 **during AOM-DSS treatment. Related to Figure 3. (A)** RT-PCR analysis for expression of
17 indicated genes in colon tissue from WT and *Card9*^{-/-} mice at indicated days after AOM injection.
18 **(B)** Densitometric analysis for the immunoblots in Figures 3C and 3E. **(C)** Frequency of CD11b⁺
19 cells and **(D)** *I18* expression in the CD11b⁺ cells from the colon, MLN and spleens of WT and
20 *Card9*^{-/-} mice on day 9. Data represent two independent experiments. Error bars represent
21 mean±standard error of the mean with 5-8 mice per group per time point.

22 **Figure S3. Cohousing normalizes disease susceptibility in WT and *Card9*^{-/-} mice. Related**
23 **to Figure 4. (A)** Percent body weight **(B)** colon length and **(C)** representative colon histology
24 pictures from WT and *Card9*^{-/-} mice housed together or separately for 4 weeks prior to AOM-
25 DSS treatment, and sacrificed on day 14. Original magnification = 10x. **(D)** Percent body weight
26 **(E)** number of tumors and **(F)** tumor distribution in the colons of WT and *Card9*^{-/-} mice housed
27 together or separately for 4 weeks prior to AOM-DSS treatment and sacrificed on day 80.
28 Principal component analysis for **(G)** 18s ITS and **(H)** 16s rRNA sequencing from the fecal
29 pellets of separately cohoused or co-housed (CH) WT and *Card9*^{-/-} mice. **(I)** Heat maps based
30 on the operational taxonomic unit (OTU) number from the 18s ITS sequencing. Data represent
31 two independent experiments and analyzed by two-way ANOVA **(A and D)** or one-way ANOVA
32 **(B and E)** followed by Holm-Sidak post-test. Error bars represent mean±standard error of the
33 mean with 5-10 mice per group per time point.

34 **Figure S4. Cohousing normalizes disease susceptibility in WT and *Card9*^{-/-} mice. Related**
35 **to Figure S3 and Figure 4.** Heat maps based on the operational taxonomic unit (OTU) number
36 in the 16s rRNA sequencing from the fecal pellets of separately cohoused or co-housed (CH)
37 WT and *Card9*^{-/-} mice with 8 mice per group.

38 **Figure S5. SYK and CARD9 are required for inflammasome activation by *Candida*.**
39 **Related to Figure 5.** Immunoblot for Caspase-1 in primary mouse bone marrow-derived
40 myeloid cells stimulated with **(A)** LPS for 4 hours or *C. albicans* for 20 hours, **(C)** LPS for 4
41 hours followed by stimulation with 5 mM ATP for 30 minutes **(D)** transfected with LPS **(E)**
42 transfected with poly(dAdT) or **(F)** stimulation with *S. Typhimurium* at MOI 1 for 2 h. **(B)** IL-18
43 levels in supernatants of cells stimulated with *Candida*. Closed arrows represent pro-caspase-1
44 (p45) and open arrows represent cleaved caspase-1 (p20). Error bars represent mean±standard
45 error of the mean.

46

47 **Figure S6. Anti-fungal treatment leads to alteration in the mycobiome. Related to Figure**
 48 **6. (A)** Principal component analysis and **(B)** heat map based on the operational taxonomic unit
 49 (OTU) number in the 18s ITS sequencing from the fecal pellets of WT mice given drinking water
 50 or amphotericin B or Itraconazole for 4 weeks. **(C)** Principal component analysis from the 16s
 51 rRNA sequencing from fecal pellets of WT mice given drinking water or amphotericin B or
 52 Itraconazole for 4 weeks. 8 mice per group.

53 **Figure S7. Metronidazole treatment leads to alteration in the gut bacterial and fungal**
 54 **landscape. Related to Figure 6.** Principal component analysis of **(A)** 16s and **(B)** 18s and **(C)**
 55 heat maps based on the operational taxonomic unit (OTU) number from the fecal pellets of WT
 56 mice given metronidazole for 5 days. 8 mice per group.

57
 58 **Supplementary Table 1. Real time qPCR primer sequences**
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Gene	Primer Sequences
<i>β-actin</i>	Forward: 5'-GGCTGTATTCCC CTCCATCG-3' Reverse: 5'-CCAGTTGGTAACAATGCCATG T-3'
<i>Muc2</i>	Forward: 5'-GCTGACGAGTGGTTGGTGAATG-3' Reverse: 5'-GATGAGGTGGCAGACAGGAGAC-3'
<i>Il22</i>	Forward: 5'-AGAACGTCTTCCAGGGTGAA-3' Reverse: 5'-CAT CGA CAT AAG TCA GCA CCA G-3'
<i>Reg3β</i>	Forward: 5'-ATGGCTCCTACTGCTATGCC-3' Reverse: 5'-GTGTCCTCCAGGCCTCTTT-3'
<i>Reg3γ</i>	Forward: 5'-ATGGCTCCTATTGCTATGC-3' Reverse: 5'-GATGTCCTGAGGGCCTCTT-3'
<i>Il22bp</i>	Forward: 5'-TCAGCAGCAAAGACAGAAGAAAC-3' Reverse: 5'-GTGTCTCCAGCCCAACTCTCA-3'
<i>Spdef</i>	Forward: 5'- AAGGCAGCATCAGGAGCAATG -3' Reverse: 5'- CTGTCAATGACGGGACACTG -3'
<i>Klf4</i>	Forward: 5'- GTGCCCCGACTAACCGTTG -3' Reverse: 5'- GTCGTTGAACTCCTCGGTCT -3'
<i>Eubacteria</i>	Forward: 5'-ACTCCTACGGGAGGCAGCAGT-3' Reverse: 5'-ATTACCGCGGCTGCTGGC-3'
<i>Bacteroides</i>	Forward: 5'-GGTTCTGAGAGGAGGTCCC-3' Reverse: 5'-CTGCCTCCCGTAGGAGT-3'
Fungal 18s	Forward: 5'- GGRAAACTC ACCAGGTCC AG -3' Reverse: 5'- GSWCTATCCCCAKCAC GA -3'
<i>Lactobacillus</i>	Forward: 5'-GGAAACAGATGCTAATACCG-3' Reverse: 5'-CACCGCTACACATGGAG-3'

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 61
 62 **Supplementary Table 2. Anti-fungal treatment leads to distinct alterations in the gut**
 63 **bacterial landscape. Related to Figure 6.** Heat map based on the operational taxonomic unit
 64 (OTU) number from the 16s rRNA sequencing from fecal pellets of WT mice given drinking
 65 water or amphotericin B for 4 weeks. 8 mice per group.

66 **Supplementary Table 3. Anti-fungal treatment leads to distinct alterations in the gut**
 67 **bacterial landscape. Related to Figure 6.** Heat map based on the operational taxonomic unit
 68 (OTU) number from the 16s rRNA sequencing from fecal pellets of WT mice given drinking
 69 water or itraconazole for 4 weeks. 8 mice per group.

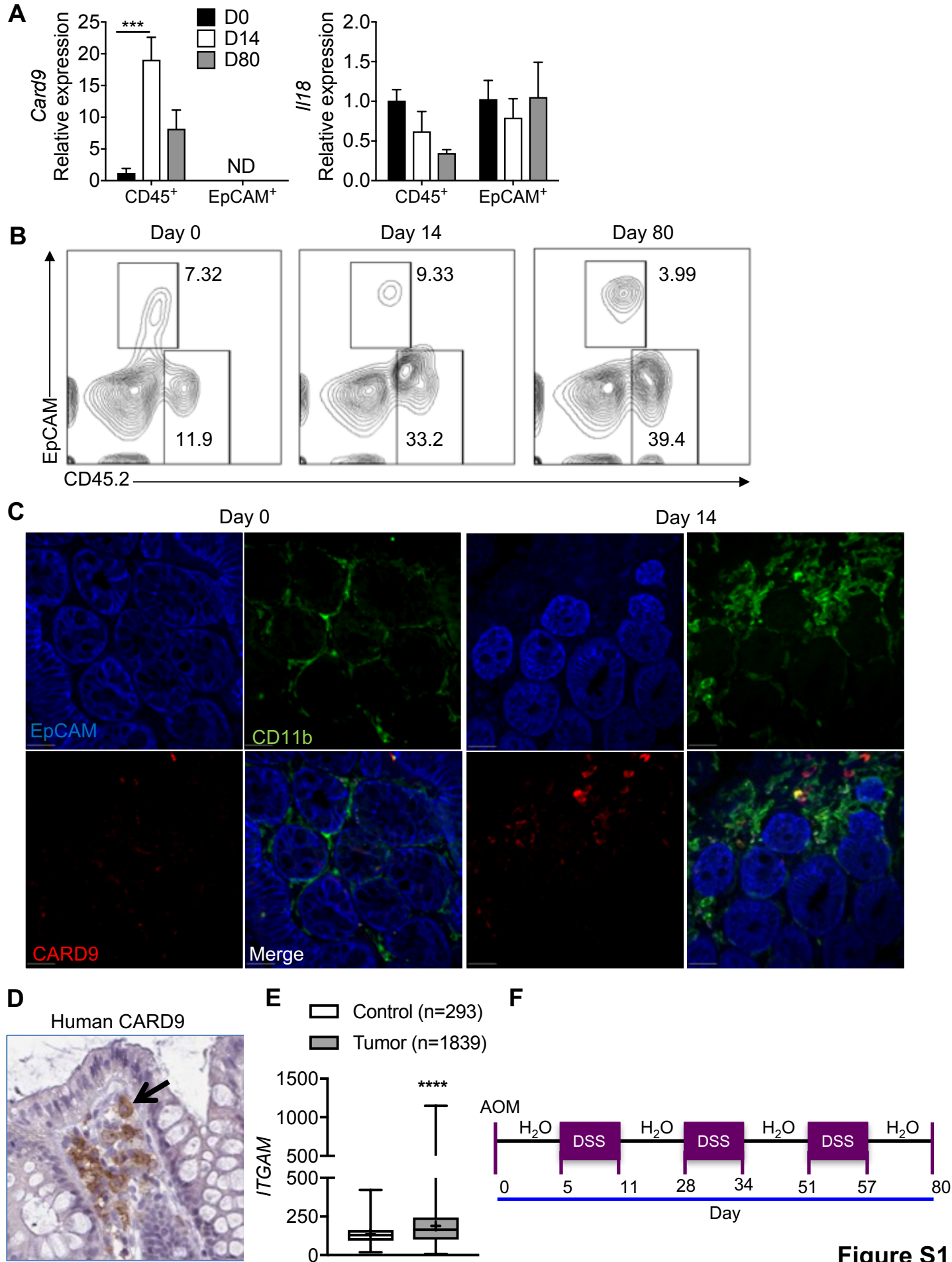


Figure S1

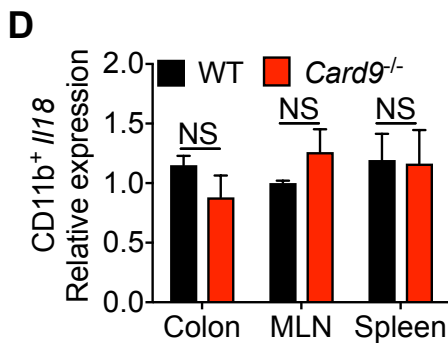
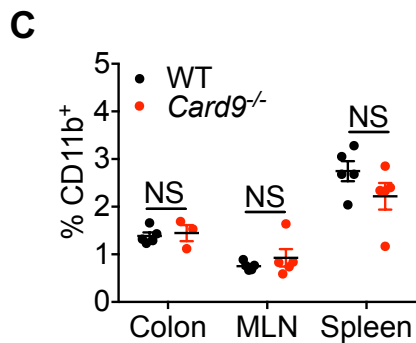
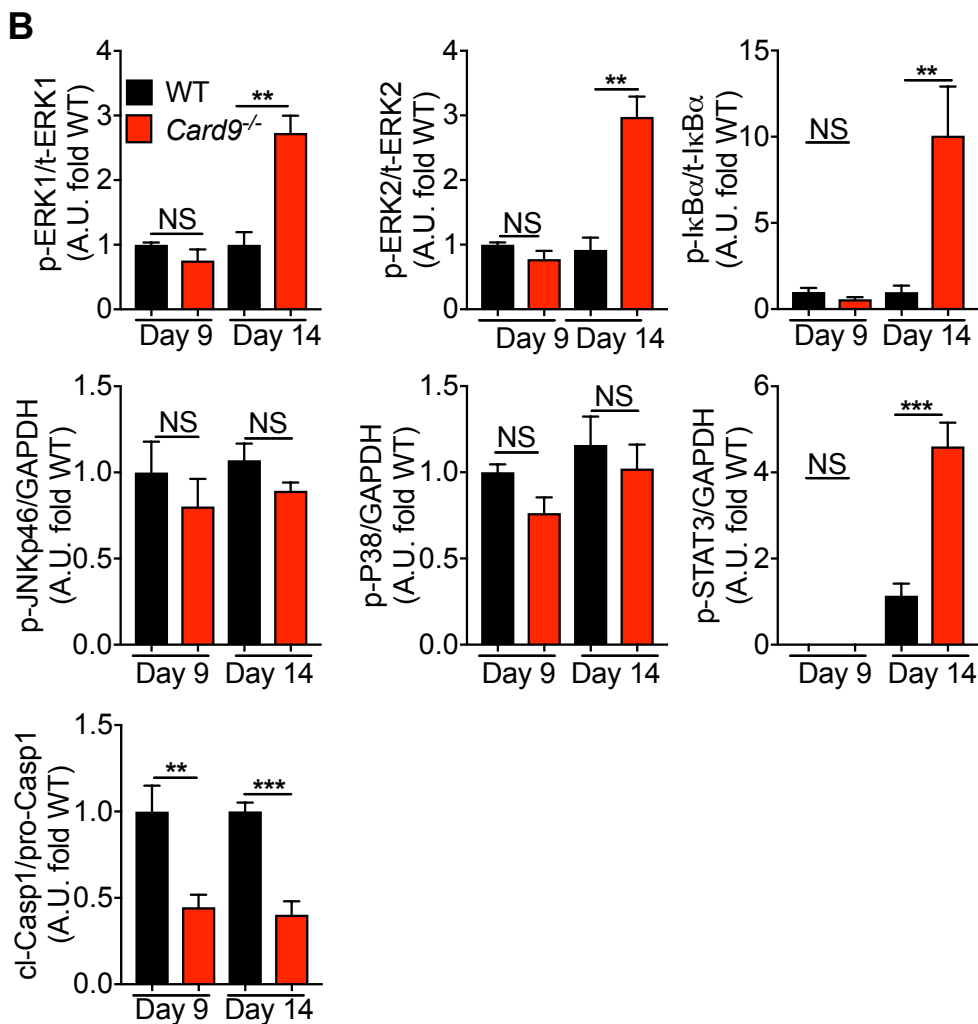
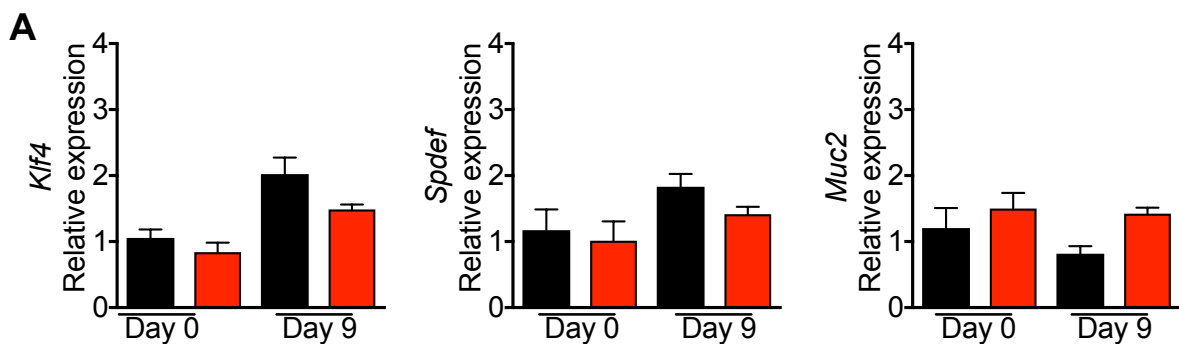


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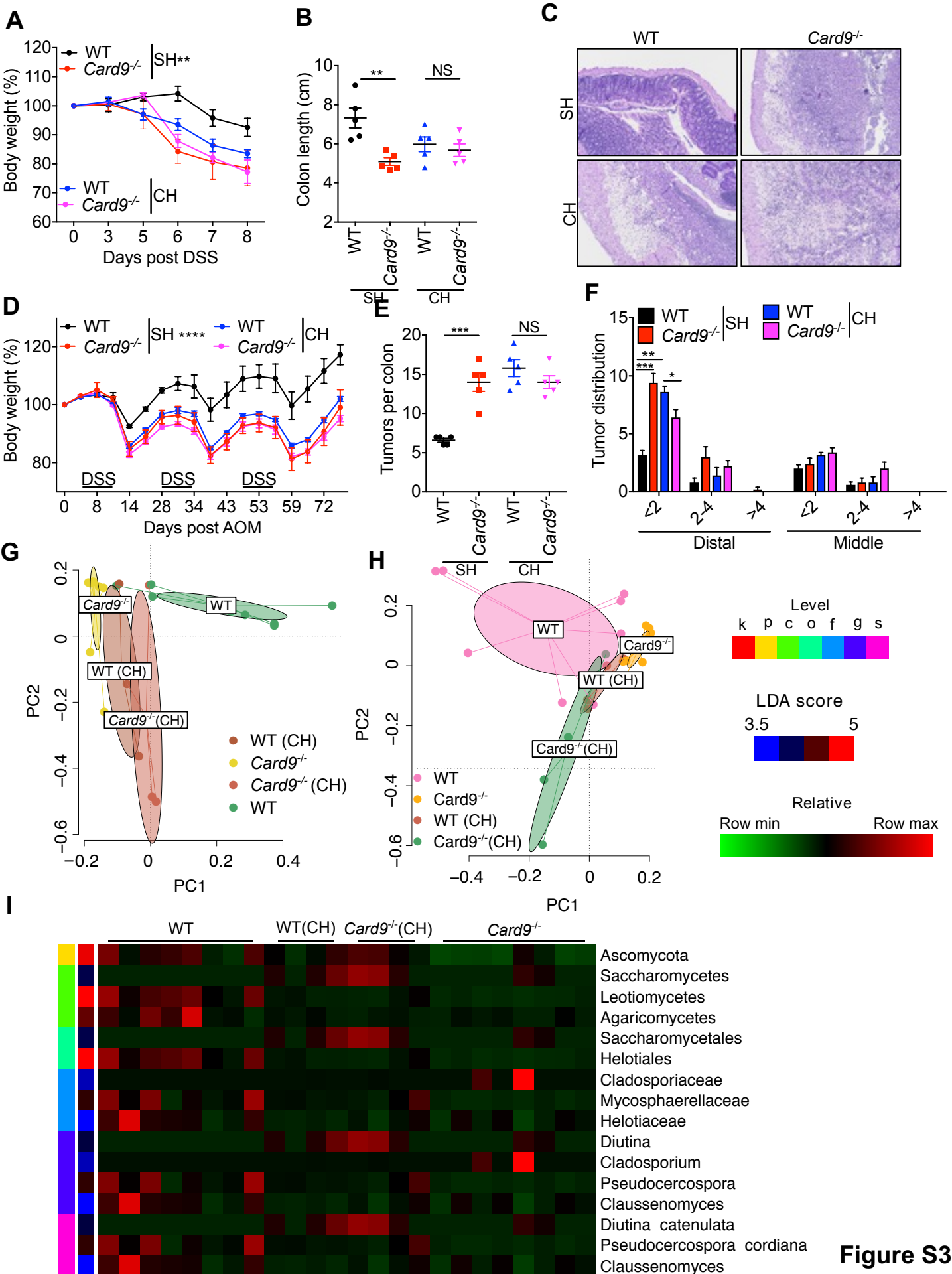
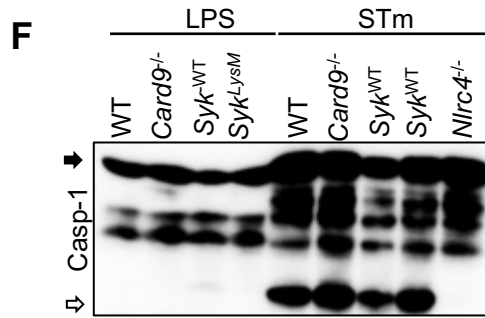
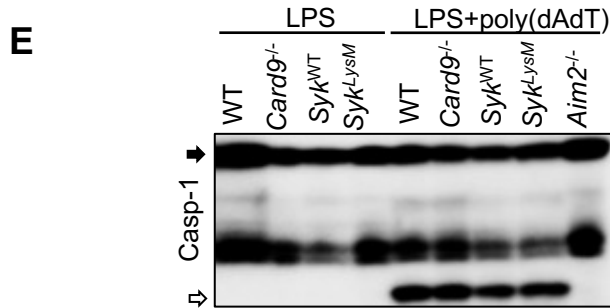
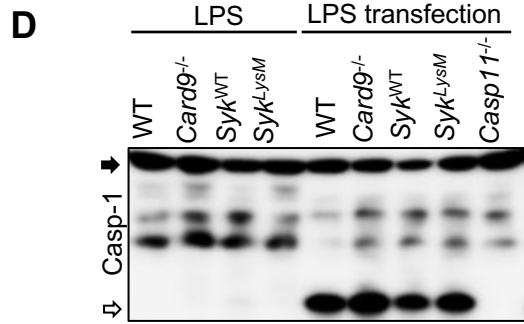
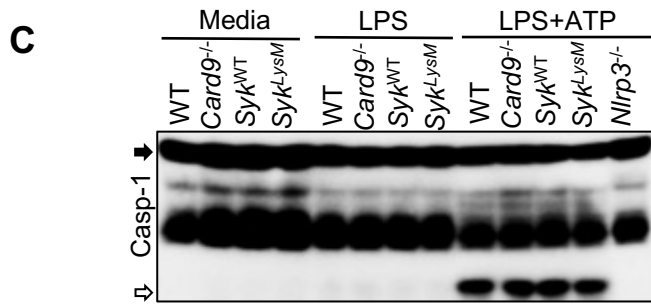
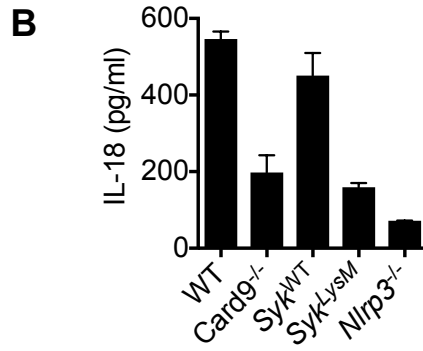
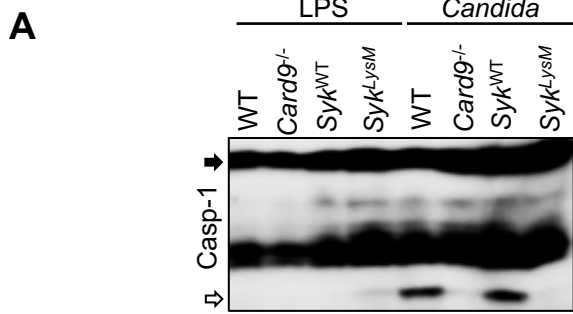


Figure S3



Figure S4



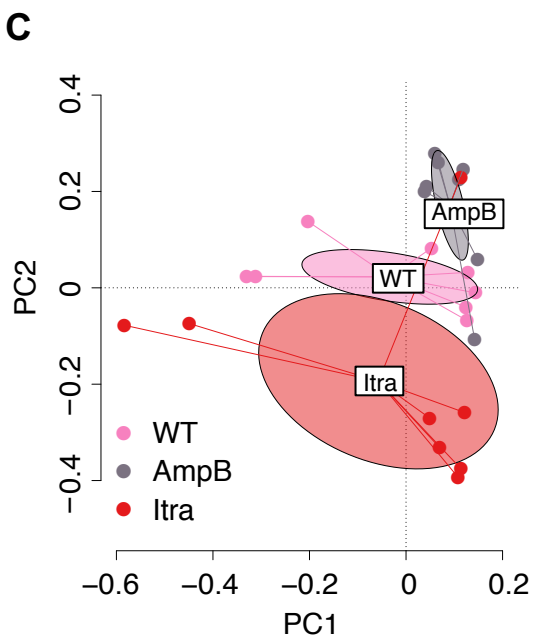
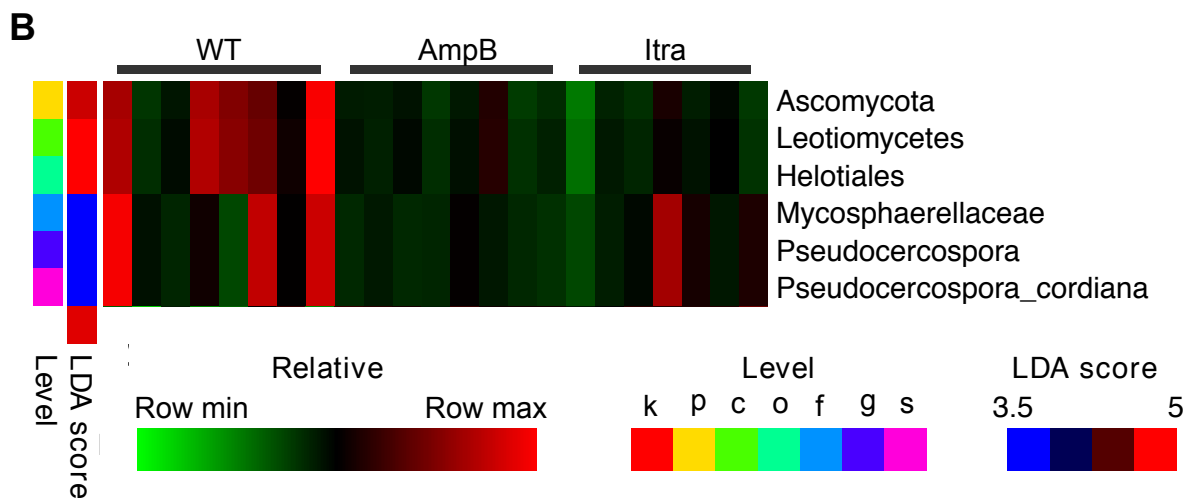
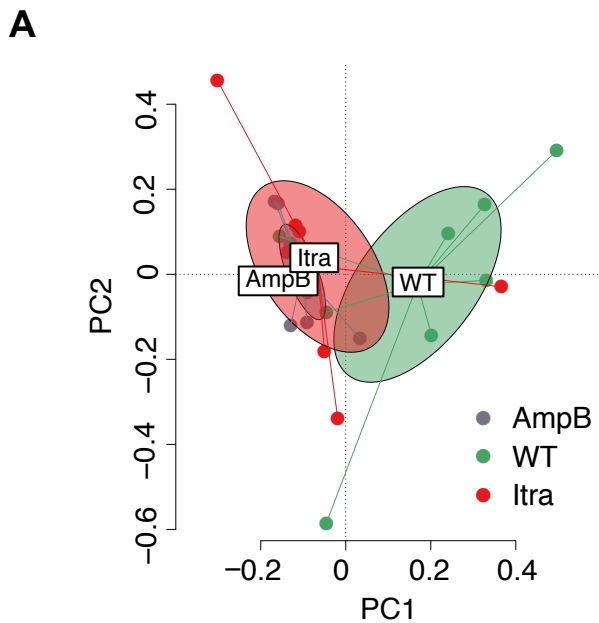


Figure S6

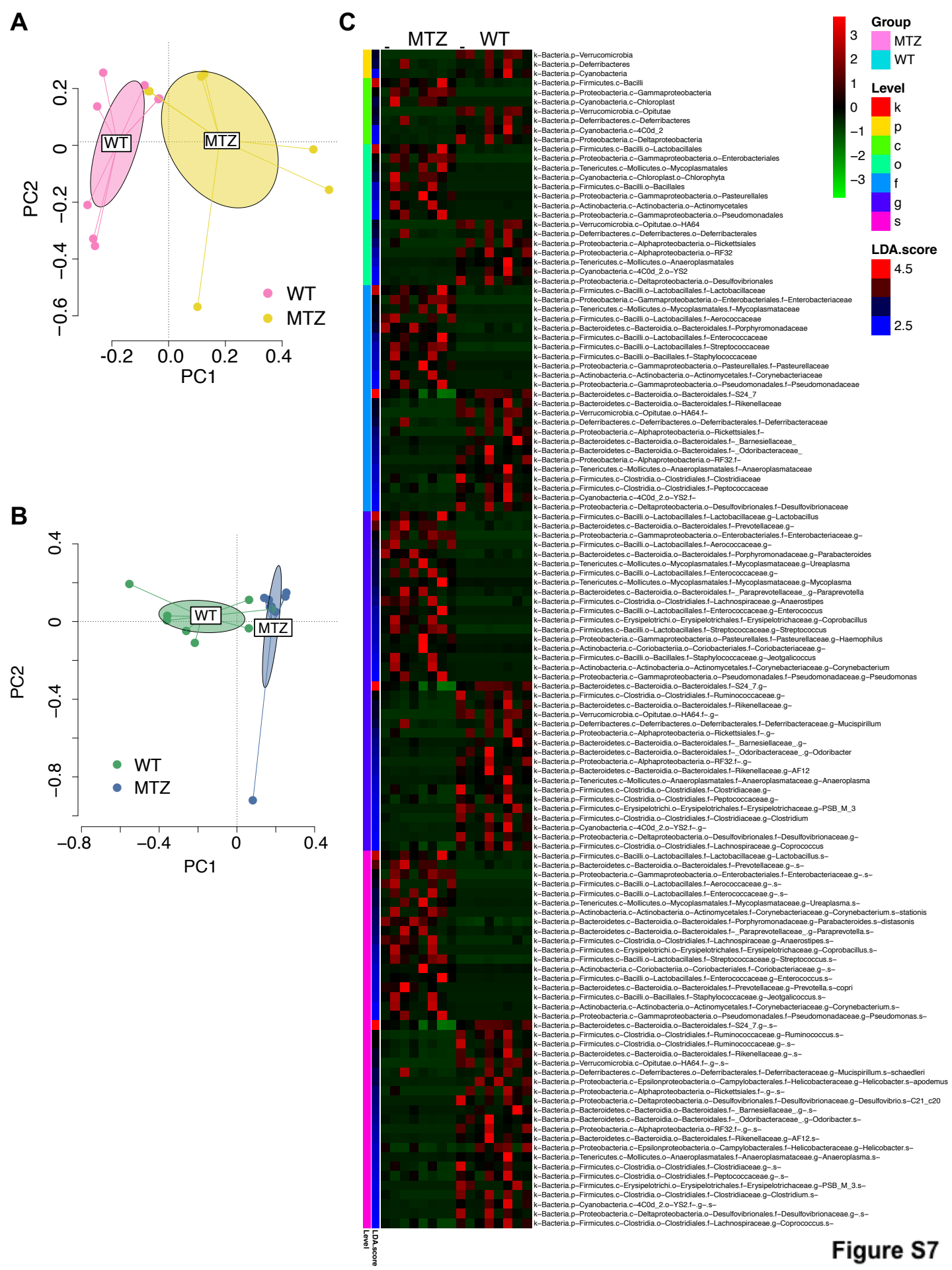


Figure S7