

Fig. S1. Relationship between bacterial communities sampled from respiratory and gut sites. Principal coordinate (PCO) plots show sample distribution in microbial community space. Plots were drawn using an weighted UniFrac distance matrix generated between samples with OTU communities rarefied to a depth of 5000 sequences per sample. The % variance explained by each principle coordinate are shown inside the plots. Nasal = nasal cavity.



Fig. S2. Shifts of bacterial community structure with age. A-D, Principal coordinate (PCO) plots. All four plots were drawn from the same weighted UniFrac distance matrix. Nasal = nasal cavity.



Fig. S3. Comparison of dynamics of core bacterial microbiota across body sites. Heat map constructed in R from log10 transformed relative abundances (abund) of the core OTUs shown in Figures 5 and 6. Sample types (left to right) were grouped using hierarchical clustering (hclust) method while bacterial phylotypes (top to bottom) were grouped according to a phylogenic tree generated using the FastTree method implemented in QIIME. 01W, 03W, etc., indicate age of birds in weeks. Nasal = nasal cavity.



Fig. S4. Change in the composition of ten genera with known pathogens of poultry over time. This figure highlights OTUs that are not highlighted in Fig. 7. Bar charts show changes in relative abundances of genus-level bacterial taxa (vertical axis) with chicken layer age (horizontal axis). 01W, 03W, etc., indicate age of birds in weeks. Nasal = nasal cavity.



Fig. S5. Antiviral and antimycoplasmal antibodies in individual birds sampled at different time points. Antiviral antibodies are presented as ELISA endpoint titers. Antimycoplasmal seroconversion is presented as the percentage of birds that tested positive over the total number of birds sampled at a given time point. AEV, avian encephalomyelitis virus (A), IBV, infectious bronchitis virus (B), IBDV, infectious bronchitis virus (C), NDV, Newcastle disease virus (D), REO, reovirus (E), MG, Mycoplasma gallisepticum and MS, M. synoviae (F). 01W, 03W, etc., indicate age of birds in weeks. Sera from 1 week old chicks had high levels of maternally derived IgG antibodies against AEV, NDV, IBDV, IBV and REO, which were metabolized and decreased to very low levels by 3 weeks of age (A-E). The maternal antibodies were due to vaccination of breeder hens. New serum IgG antibodies emerged and increased in titers depending on the timing of the vaccination regimens (for AEV, IBDV, NDV, and IBV) or the onset of infection (for REO). The wide range of antibody titers enabled the analysis of differential abundance of specific bacterial taxa as described in other sections in this paper. Antibody titers are not available for MG and MS because the tests used for antibody detection were qualitative. All the same, only less than 20% of sampled chickens were seropositive for the MG vaccine from 25 weeks onward (F). The first serological evidence of MS infection was at 36 weeks and more than 50% of birds were positive at 51 weeks (F).

Fig. S6.



Fig. S6. Sampling time-points, feed changes, and vaccination/antibiotic treatment history. Brood stage = 0-5 weeks, Grow-out stage = 6 -16 weeks, laying stage = 17 weeks and older. Bursimune (Ceva Animal Health, USA) and Bursine-2 (Zoetis, USA) vaccines contain live infectious bursal disease virus. Avipro IB Ark (Elanco Animal Health, USA) is a vaccine consisting of the infectious bronchitis virus (Ark type), Avipro ND IB Sohol (Elanco Animal Health, USA) is an attenuated live vaccine consisting of the Newcastle disease virus (B1 type, LaSota strain) and infectious bronchitis virus (Mass type). AE-Poxine (Zoetis, USA) vaccine contains live attenuated avian encephalomyelitis and fowl pox viruses. Mildvac-Mass+Conn (MERCK animal health, USA) is a live attenuated vaccine against Massachusetts and Connecticut type infectious bronchitis viruses. Laryngo-vac (Zoetis, USA) is a modified live vaccine against infectious laryngotracheitis virus. Mycovac-L (MERCK animal health, USA) contains live attenuated 6/85 strain of Mycoplasma gallisepticum. Pox Blen (Merial, USA) is a pigeon Pox (Hitchner strain) live virus CEO vaccine against fowl and pigeon pox viruses. BMD 50 (Zoetis, USA) is an antibiotic used to control bacterial enteritis.