

Food supplementation affects gut microbiota and immunological resistance to parasites in a wild bird species

S.A. Knutie

Table S1. Pairwise correlation coefficients between growth metrics (bill length, tarsus length, first primary feather length, and body mass).

	Bill length	Tarsus length	Feather length	Body mass
Bill length	1.00			
Tarsus length	0.37	1.00		
Feather length	0.65	0.47	1.00	
Body mass	0.48	0.38	0.66	1.00

Table S2. The effect of parasite treatment, food treatment, and their interaction on parasite abundance.

Variables	Coefficients \pm SE
Parasite treatment	12.89 \pm 10.60
Food treatment	0.00 \pm 10.60
Parasite x food treatment	42.66 \pm 13.94

Table S3. The effect of food treatment, Julian date, and their interaction on parasite abundance within the sham-fumigated treatment.

Variables	Coefficients \pm SE
Food treatment	21.24 \pm 8.88
Julian date	0.05 \pm 0.04
Food treatment x Julian date	-0.11 \pm 0.05

Table S4. The effect of parasite treatment, food treatment, and their interaction on growth, hemoglobin levels, and glucose levels.

Variables	Coefficients \pm SE
Bill length	
Parasite treatment	0.00 \pm 0.02
Food treatment	-0.02 \pm 0.02
Parasite x food treatment	0.01 \pm 0.02
Tarsus	
Parasite treatment	0.02 \pm 0.01
Food treatment	0.00 \pm 0.01
Parasite x food treatment	-0.01 \pm 0.01
First primary length	
Parasite treatment	0.02 \pm 0.06
Food treatment	-0.10 \pm 0.07
Parasite x food treatment	0.05 \pm 0.08
Body mass	
Parasite treatment	-0.00 \pm 0.02
Food treatment	-0.03 \pm 0.02
Parasite x food treatment	-0.00 \pm 0.02
Hemoglobin	
Parasite treatment	-0.05 \pm 0.06
Food treatment	-0.01 \pm 0.07
Parasite x food treatment	-0.10 \pm 0.09
Glucose	
Parasite treatment	-0.04 \pm 0.08
Food treatment	-0.05 \pm 0.09
Parasite x food treatment	0.01 \pm 0.12

Table S5. The effect of food treatment on immune responses and bacterial diversity metrics.

Variables	Coefficients \pm SE
IgY antibody levels	-0.28 \pm 0.11
Haptoglobin levels	-0.03 \pm 0.07
Bacterial diversity	
sobs	-0.30 \pm 0.17
Shannon index	-1.06 \pm 0.74
Simpson Index	-0.32 \pm 0.27

Table S6. The effect of food treatment on mean \pm SE bacterial diversity metrics. Numbers in the parentheses are the sample sizes of nests.

Variables	Supplemented	Unsupplemented
sobs	343.80 \pm 56.04 (7)	204.25 \pm 51.28 (6)
Shannon index	3.14 \pm 0.41 (7)	2.06 \pm 0.64 (6)
Simpson index	12.85 \pm 3.38 (7)	9.60 \pm 5.00 (6)

Table S7. Relative abundances (%) of bacterial genera that differ between the birds (supplemented: S, unsupplemented: US) and mealworms (MW). Numbers (%) in bold indicate which treatment of birds are statistically different from the mealworms (Tukey: $P < 0.05$).

Genus	MW	S	US	Statistics
<i>Brachybacterium</i>	1.90	0.08	0.00	$F = 67.12, P < 0.0001$
<i>Brevibacterium</i>	5.08	0.13	0.00	$F = 103.35, P < 0.0001$
<i>Candidatus</i>	0.00	2.68	2.41	$F = 18.38, P = 0.002$
<i>Dietzia</i>	1.25	0.00	0.00	$F = 11.53, P = 0.01$
<i>Erysipelatoclostridium</i>	0.00	2.46	3.89	$F = 9.17, P = 0.03$
<i>Hyphomicrobium</i>	0.00	1.43	0.97	$F = 8.14, P = 0.04$
<i>Kocuria</i>	1.90	0.20	0.00	$F = 34.96, P = 0.0001$
<i>Lactobacillus</i>	8.96	0.45	0.00	$F = 31.82, P = 0.0001$
<i>Lactococcus</i>	15.88	0.72	0.67	$F = 31.91, P = 0.0001$
<i>Methylobacterium</i>	0.62	3.83	3.94	$F = 9.27, P = 0.03$
<i>Mycobacterium</i>	0.00	2.53	2.50	$F = 11.55, P = 0.01$
<i>Nocardioides</i>	0.00	6.01	5.93	$F = 24.47, P < 0.001$
<i>Pantoea</i>	1.90	0.66	0.66	$F = 7.57, P = 0.04$
<i>Parabacteroides</i>	1.27	0.08	0.00	$F = 7.56, P = 0.04$
<i>Parasutterella</i>	1.90	0.00	0.00	$F = 40015.68, P < 0.0001$
<i>Rhizobium</i>	0.00	1.40	1.67	$F = 9.85, P = 0.02$
<i>Rhodococcus</i>	0.00	2.32	2.13	$F = 9.77, P = 0.02$
<i>Spiroplasma</i>	10.78	0.33	0.20	$F = 64.65, P < 0.0001$
<i>Staphylococcus</i>	5.07	0.52	0.35	$F = 11.96, P = 0.01$
<i>Variibacter</i>	0.00	0.97	0.56	$F = 7.75, P = 0.04$
<i>Weissella</i>	9.41	0.22	0.20	$F = 35.30, P = 0.0001$
<i>Xanthomonas</i>	1.90	0.08	0.18	$F = 18.99, P = 0.001$



Fig. S1. Experimental mealworm feeder being used by a male eastern bluebird.