Supporting Information

Sharon et al. 10.1073/pnas.1009906107

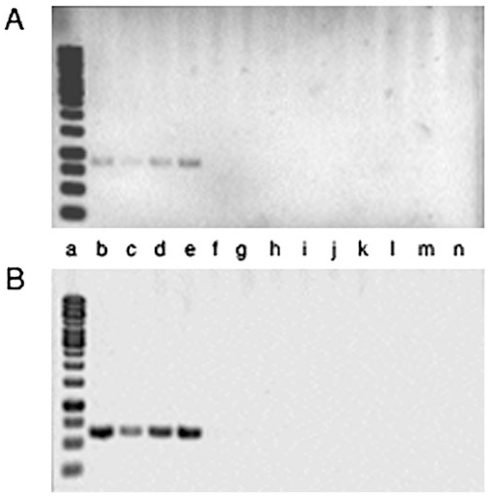

Fig. S1. PCR analysis for *Wolbachia*. Electrophoresis in 1% agarose of *Wolbachia*-specific 16S rRNA gene (*A*) and *wsp* gene PCR products (*B*). Lane a, Fermentas GeneRuler 1 kb DNA ladder (three shortest bands are 750 bp, 500 bp, and 250 bp); lanes b–e, male and female 20th-generation flies, reared on CMY (lanes b and c, CMY; lanes d and e, starch); lanes f–i, male and female 20th-generation flies, reared on CMY supplemented with a mixture of antibiotics (lanes f and g, CMY; lanes h and i, starch); lanes j–m, male and female 20th-generation flies, reared on CMY supplemented with their respective bacteria (infection experiment) after treatment with a mixture of antibiotics (lanes j and k, CMY; lanes l and m, starch).

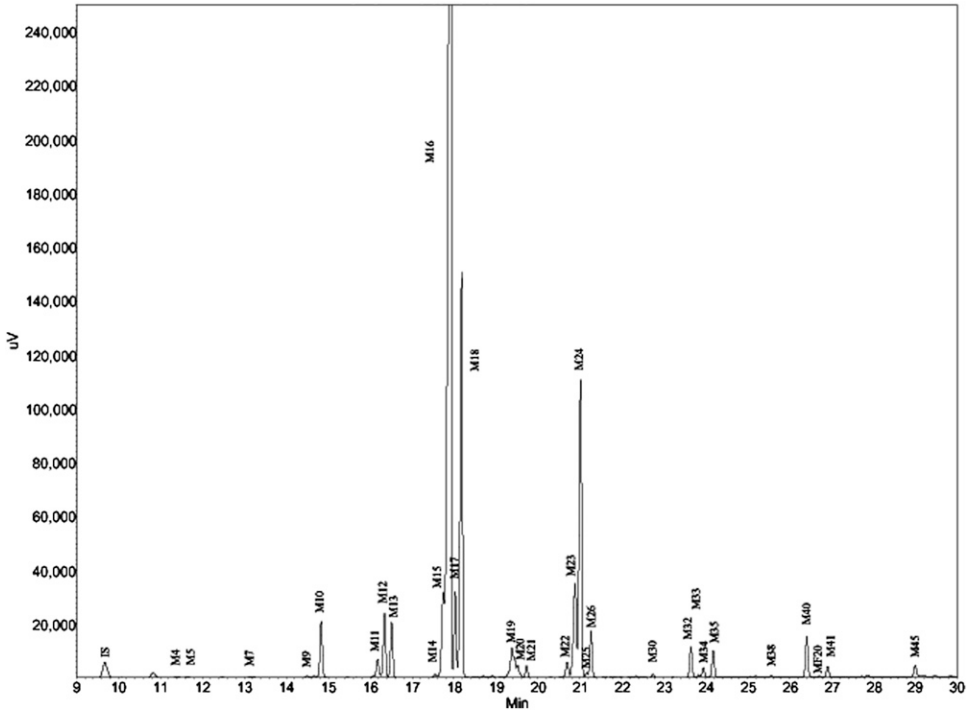

Fig. S2. GC-FID chromatogram of pentane-extracted CHs of CMY male flies (n = 10). Peak annotations correspond to peak names in Table S1.

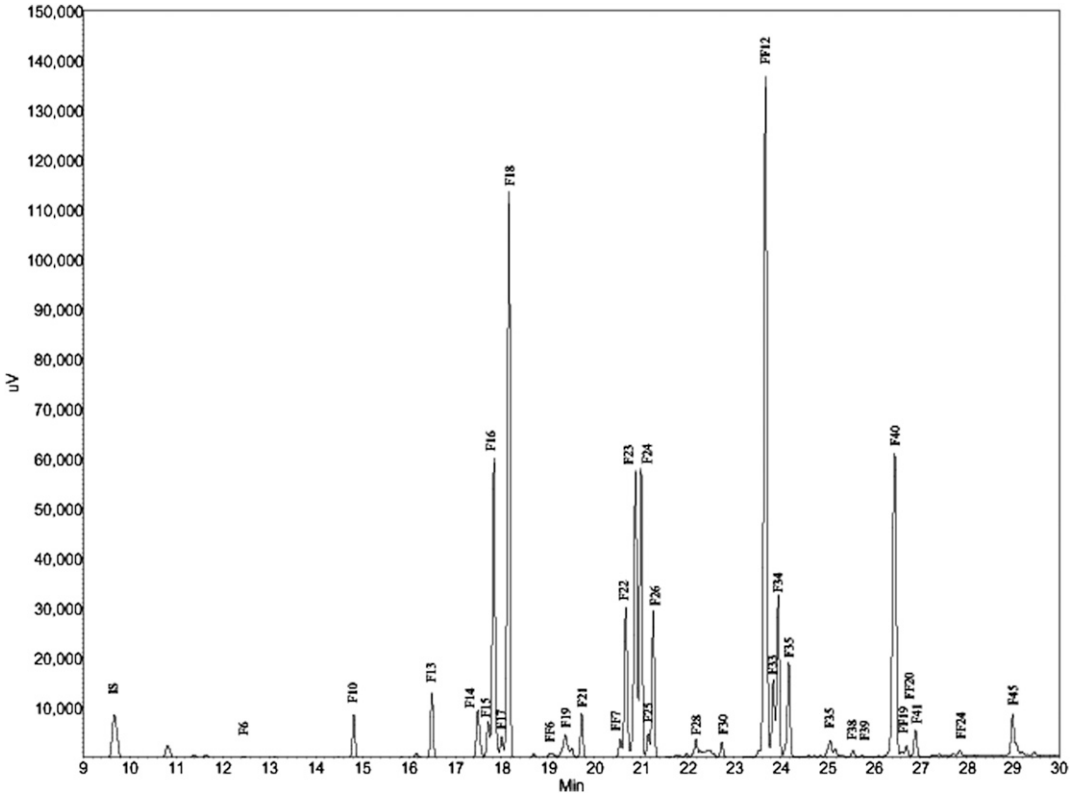

Fig. S3. GC-FID chromatogram of pentane-extracted CHs of CMY female flies (n = 9). Peak annotations correspond to peak names in Table S2.

Table S1. CH profiles of untreated and antibiotic-treated CMY- and starch-bred male flies

Peak name	Retention time (min)	Presumed compound*	Mean CH per three flies ± SEM, ng			
			No antibiotic treatment		Treated with antibiotics	
			CMY male (n = 3)	Starch male (n = 3)	CMY male (n = 3)	Starch male (n = 3)
M1	4.36	—	16.0 ± 6.1	21.8 ± 4.2	29.5 ± 3.4	42.9 ± 10.2
M2	7.2	—	2.6 ± 1.4	9.4 ± 2.8	1.8 ± 0.9	3.9 ± 1.5
M3	8.12	—	6.7 ± 0.3	7.0 ± 0.6	5.3 ± 0.3	7.0 ± 0.7
IS	9.75	c-18 internal standard	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0
M4	11.46	—	2.1 ± 0.4	3.1 ± 0.3	ND	ND
M5	11.71	—	3.3 ± 0.6	3 ± 0.3	3.3 ± 0.4	3.2 ± 0.1
M7	13.26	—	1.6 ± 1.1	3.9 ± 1.6	ND	ND
M9	14.53	—	2.0 ± 2.0	7.8 ± 2.7	ND	ND
M10	14.89	—	27.3 ± 4.6	64.1 ± 5.3	19.3 ± 1.6	53.4 ± 3.8
M11	16.24	9-Docosene†	7.4 ± 1.5	17.0 ± 1.1	6.5 ± 0.6	16.9 ± 0.2
M12	16.4	cis-vaccenyl acetate†	29.8 ± 18.6	154.8 ± 23.5	48.5 ± 17.0	97.1 ± 42.1
M13	16.57	—	37.3 ± 7.3	55.3 ± 5.8	27.2 ± 2.1	38.1 ± 0.5
M14	17.6	2-Methyldocosane	2.1 ± 0.4	8.7 ± 1.3	1.5 ± 0.2	4.2 ± 0.5
M15	17.78	9-Tricosene†	32.8 ± 6.3	22.4 ± 5.2	32.1 ± 7.2	48.7 ± 3.2
M16	17.92	7-Tricosene†	777.3 ± 177.1	1486.2 ± 119.7	543.6 ± 47.5	1245.0 ± 31.6
M17	18.07	5-Tricosene†	45.2 ± 9.2	67.7 ± 3.7	27.0 ± 1.0	58.3 ± 4.0
M18	18.22	n-tricosane†	345.8 ± 53.4	385.2 ± 48.2	196.8 ± 21.3	277.9 ± 28.6
M19	19.45	7-Tetracosene	29.3 ± 6.0	26.8 ± 3.8	16.5 ± 6.5	26.7 ± 0.5
M20	19.6	5-Tetracosene	2.4 ± 0.6	3.8 ± 0.5	1.9 ± 0.4	5.9 ± 4.4
M21	19.8	n-tetracosane	13.3 ± 2.2	13.1 ± 2.6	7.2 ± 1.1	6.8 ± 0.4
M22	20.79	2-Methyltetracosane	12.2 ± 3.1	42.9 ± 6.1	5.9 ± 0.9	21.7 ± 1.9
M23	20.98	9-Pentacosene†	108.0 ± 21.7	63.1 ± 16.4	85.8 ± 3.7	60.3 ± 3.4
M24	21.11	7-Pentacosene†	437.9 ± 89.4	184.5 ± 25.1	279.3 ± 10.6	209.0 ± 14.3
M25	21.27	5-Pntacosene†	4.8 ± 0.9	0.9 ± 0.5	3.4 ± 1.0	1.4 ± 0.1
M26	21.38	n-pentacosane†	50.6 ± 7.9	49.8 ± 12.2	18.1 ± 5.0	25.2 ± 4.0
M30	22.9	2-Methylpentacosane	5.5 ± 1.0	3.7 ± 1.1	ND	ND
M32	23.67	2-Methylhexacosane†	15.4 ± 15.4	87.6 ± 31.6	12.6 ± 1.6	30.8 ± 1.3
M33	24.03	—	3.7 ± 0.6	6.2 ± 4.6	ND	ND
M34	24.16	7-Heptacosene	39.2 ± 10.0	9.8 ± 6.4	19.9 ± 3.3	5.1 ± 0.7
M35	24.4	n-heptacosane†	58.7 ± 10.4	24.6 ± 6.7	13.4 ± 3.9	13.2 ± 3.2
M38	25.83	—	5.6 ± 1.2	1.4 ± 0.3	ND	ND
M40	26.72	2-Methyloctacosane†	77.5 ± 16.6	70.7 ± 21.5	15.5 ± 3.9	26.0 ± 3.0
MF20	27.02	—	3.4 ± 0.9	0.8 ± 0.4	ND	ND
M41	27.24	n-nonacosane	46.0 ± 12.2	7.0 ± 1.1	6.8 ± 1.7	4.7 ± 1.5
MF24	28.24	—	4.6 ± 2.3	1.1 ± 0.3	ND	ND
M43	28.59	—	3.4 ± 1.0	1.3 ± 1.2	ND	ND
M45	29.42	2-Methyltriacontane	35.3 ± 6.6	9.1 ± 1.3	5.5 ± 1.6	4.9 ± 0.8
M46	29.91	—	3.7 ± 2.3	0.4 ± 0.2	ND	ND
Total amount			2,347.9 ± 468.1	2,948.1 ± 335.3	1,453.4 ± 83.2	2,360.4 ± 130.4

CHs that were less than 3 ng per sample were omitted from this table. ND, not detectable.
*Based on the GC CH profiles of *D. melanogaster* (1) and on GC-MS analyses.
†Identified by GC-MS.

1. Everaerts C, Farine JP, Cobb M, Ferveur JF (2010) Drosophila cuticular hydrocarbons revisited: Mating status alters cuticular profiles. *PLoS ONE* 5:e9607.

Table S2. CH profiles of untreated and antibiotic-treated CMY and starch bred female flies

			Mean CH per three flies ± SEM, ng			
			No antibiotic treatment		Treated with antibiotics	
Peak name	Retention time (min)	Presumed compound*	CMY female (n = 3)	Starch female (n = 2)	CMY female (n = 3)	Starch female (n = 3)
F1	4.1	—	19.6 ± 4.3	9.8 ± 3.6	32.6 ± 4.0	25.9 ± 6.7
F2	7.02	—	6.1 ± 0.7	5.4 ± 1.6	3.6 ± 1.3	4.5 ± 2.6
F3	8.04	—	7.0 ± 1.3	4.6 ± 1.2	5.7 ± 0.6	6.7 ± 0.9
IS	9.7	c-18 internal standard	100 ± 0.0	100 ± 0.0	100 ± 0.0	100 ± 0.0
F5	11.66	—	ND	ND	3.1 ± 0.2	3.6 ± 0.3
F6	12.5	—	3.3 ± 0.5	2.8 ± 0.0	ND	ND
F8	13.58	—	0.7 ± 0.1	1.3 ± 0.5	ND	ND
F10	14.89	n-heneicosane	17.3 ± 3.8	16.6 ± 0.9	8.5 ± 0.4	9.4 ± 0.1
F13	16.57	n-docosene	29.8 ± 4.2	25.3 ± 1.2	15.4 ± 0.3	13.4 ± 1.7
F14	17.56	2-Methyldocosane	16.8 ± 2.4	25.2 ± 0.3	11.1 ± 1.6	16.8 ± 1.8
F15	17.78	9-Tricosene†	8.8 ± 2.0	5.0 ± 0.4	6.7 ± 0.4	6.8 ± 0.5
F16	17.92	7-Tricosene†	134.1 ± 30.4	67.9 ± 1.6	77.3 ± 6.9	48.7 ± 2.4
F17	18.07	5-Tricosene†	7.3 ± 2.6	2.2 ± 0.1	3.7 ± 0.5	1.8 ± 0.3
F18	18.24	n-tricosane†	220.8 ± 35.0	222.5 ± 10.6	122.8 ± 7.6	112.7 ± 9.0
FF6	19.16	7,11-Tetracosadiene	2.1 ± 1.1	3.5 ± 0.8	ND	ND
F19	19.44	7-Tetracosene	13.0 ± 1.1	7.5 ± 1.4	9.0 ± 0.6	6.4 ± 0.6
F21	19.79	n-tetracosane	20.1 ± 01.0	16.8 ± 1.5	12.3 ± 0.5	11.0 ± 1.9
FF7	20.62	9,13-Pentacosadiene	4.7 ± 0.7	10.2 ± 1.1	2.9 ± 0.2	4.3 ± 1.0
F22	20.75	7,11-Pentacosadiene	74.7 ± 24.0	70.0 ± 0.4	26.6 ± 3.2	30.4 ± 2.1
F23	20.96	9-Pentacosene†	128.1 ± 16.0	114.5 ± 17.3	89.0 ± 6.3	70.3 ± 4.1
F24	21.08	7-Pentacosene†	111.3 ± 23.2	33.2 ± 6.8	60.9 ± 7.4	31.6 ± 2.3
F25	21.22	5-Pentacosene†	4.1 ± 0.8	2.2 ± 0.4	3.4 ± 0.4	1.6 ± 0.0
F26	21.34	n-pentacosane†	51.6 ± 4.4	49.7 ± 2.8	33.0 ± 3.3	34.2 ± 3.1
F28	22.25	7,11-Hexacosadiene	6.7 ± 1.2	8.6 ± 0.6	3.2 ± 0.5	3.5 ± 0.5
F30	22.81	2-Methylpentacosane	5.8 ± 3.1	8.1 ± 0.7	4.5 ± 0.2	4.2 ± 0.3
FF12	23.77	7,11-Heptacosadiene†	352.1 ± 65.5	516.8 ± 16.7	151.9 ± 24.3	178.3 ± 26.4
F33	23.92	9-Heptacosene†	22.4 ± 0.5	36.7 ± 2.9	19.4 ± 1.9	20.0 ± 2.8
F34	24.03	7-Heptacosene†	57.7 ± 3.7	21.8 ± 5.4	44.5 ± 3.5	28.1 ± 1.0
F35	24.26	n-heptacosane†	68.2 ± 14.9	69.6 ± 4.1	26.5 ± 1.3	26.1 ± 0.7
F36	25.14	7,11-Nonacosadiene	10.2 ± 0.4	15.4 ± 1.5	4.1 ± 0.4	5.5 ± 0.6
F38	25.64	—	7.5 ± 1.7	6.7 ± 0.6	ND	ND
F39	25.83	—	1.6 ± 0.2	2.2 ± 0.2	ND	ND
F40	26.54	2-Methyloctacosane†	261.5 ± 7.5	408.5 ± 12.9	102.7 ± 13.0	146.9 ± 18.2
FF19	26.68	—	2.3 ± 0.2	3.4 ± 0.5	ND	ND
FF20	26.79	7-Nonacosene	7.4 ± 0.6	4.1 ± 0.6	3.6 ± 0.2	4.3 ± 1.9
F41	26.99	n-nonacosane†	39.6 ± 12.6	37.9 ± 2.9	9.3 ± 0.7	7.0 ± 1.7
FF24	27.93	—	5.4 ± 1.8	3.1 ± 0.4	ND	ND
F45	29.07	2-Methyltriacontane	44.1 ± 11.7	49.1 ± 8.6	13.2 ± 1.7	11.6 ± 2.7
FF26	29.28	—	1.8 ± 0.1	2.2 ± 0.1	1.0 ± 0.1	2.2 ± 1.6
F46	29.53	—	3.7 ± 1.2	3.0 ± 0.1	ND	ND
Total amount			1,753.9 ± 258.7	1,926.8 ± 82.4	937.2 ± 83.4	915.3 ± 49.2

Cuticular hydroocarbons that were less than 3 ng per sample were omitted from this table. ND, not detectable.
*Based on the GC CH profiles of *D. melanogaster* (1) and on GC-MS analyses.
†Identified by GC-MS.

1. Everaerts C, Farine JP, Cobb M, Ferveur JF (2010) Drosophila cuticular hydrocarbons revisited: Mating status alters cuticular profiles. *PLoS ONE* 5:e9607.