

Genetic basis of octanoic acid resistance in *Drosophila sechellia*: functional analysis of a fine-mapped region

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Supplemental Figure legends

Figure S1: Measuring OA associated survivorship curves. Survival curves from the OA resistance assay are shown (**A**) all lines of *D. sechellia* (red), *D. melanogaster* (blue), *D. mauritiana* (green), and *D. simulans* (orange) (for relative survival see Figure 1C) with data from the sexes combined and (**B**) for species means separated by sex with females indicated by a darker shade. Dotted lines in (**B**) represent 95% confidence intervals from a Cox regression model.

Figure S2: Relative survival of mutant line associated with OA resistance. Relative survival ($-\beta$) from a Cox regression model comparing line carrying mutant allele of *Obp83g* to baseline of the background in which this mutant line was made, see Table S1. Error bars represent 2SE.

Figure S3: Osiris candidate gene expression profiles. **A)** Gene expression levels from a previous study show developmental expression patterns in *D. melanogaster* (Graveley *et al.* 2012) (*y[1]*; *cn[1]* *bw[1]* *sp[1]*) for *Osi6* (black bars), *Osi7* (grey bars) and *Osi8* (white bars). **B)** Gene expression levels from a previous study reported tissue-specific expression profiles in *D. melanogaster* (Oregon R) (Brown *et al.* 2014) for *Osi6* (black bars), *Osi7* (grey bars) and *Osi8* (white bars). **C)** Gene expression levels from a previous study reported induction in *D. melanogaster* (Oregon R) in response to perturbations (Brown *et al.* 2014) for *Osi6* (black bars), *Osi7* (grey bars) and *Osi8* (white bars).

Figure S4: Relative survival of tissue-specific RNAi knockdown of *Osi5* in response to OA exposure. Relative survival ($-\beta$) comparing tissue-specific knockdown (tissue-specific GAL4/UAS-RNAi) to a baseline of UAS-RNAi. Both tissue-specific knockdown (black) and parental control (tissue-specific GAL4, grey) are shown. Error bars represent 2SE.

Supplemental Table legends

Table S1: *Drosophila melanogaster* RNAi, GAL4 and mutant lines used in this study. For each, the gene affected, stock number, genotype and source is indicated.

Table S2: Sample sizes for OA resistance experiments of different *Drosophila* species. For each tested line, the species, strain and sample size are indicated.

Table S3: Sample sizes for OA resistance experiments tested by RNAi. For each gene in the mapped resistance region, sample sizes for ubiquitous knockdown are shown separated by UAS-RNAi line only, knockdown GAL4XUAS-RNAi, and sibling controls (GAL4XUAS-RNAi CyO). For stage-specific RNAi resistance assays, sample sizes are shown for hormone induced knockdown (H) and uninduced siblings.

Table S4: Primers used in qRT-PCR measurement of gene expression.

Figure S1

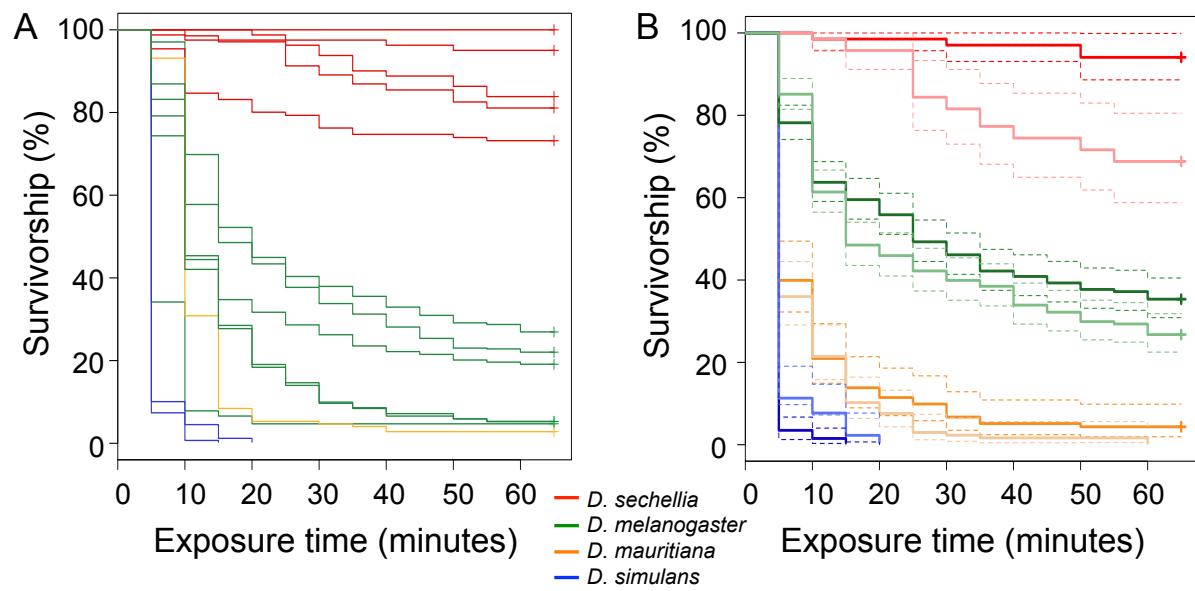


Figure S2

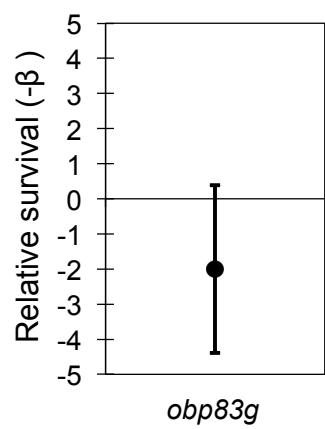


Figure S3

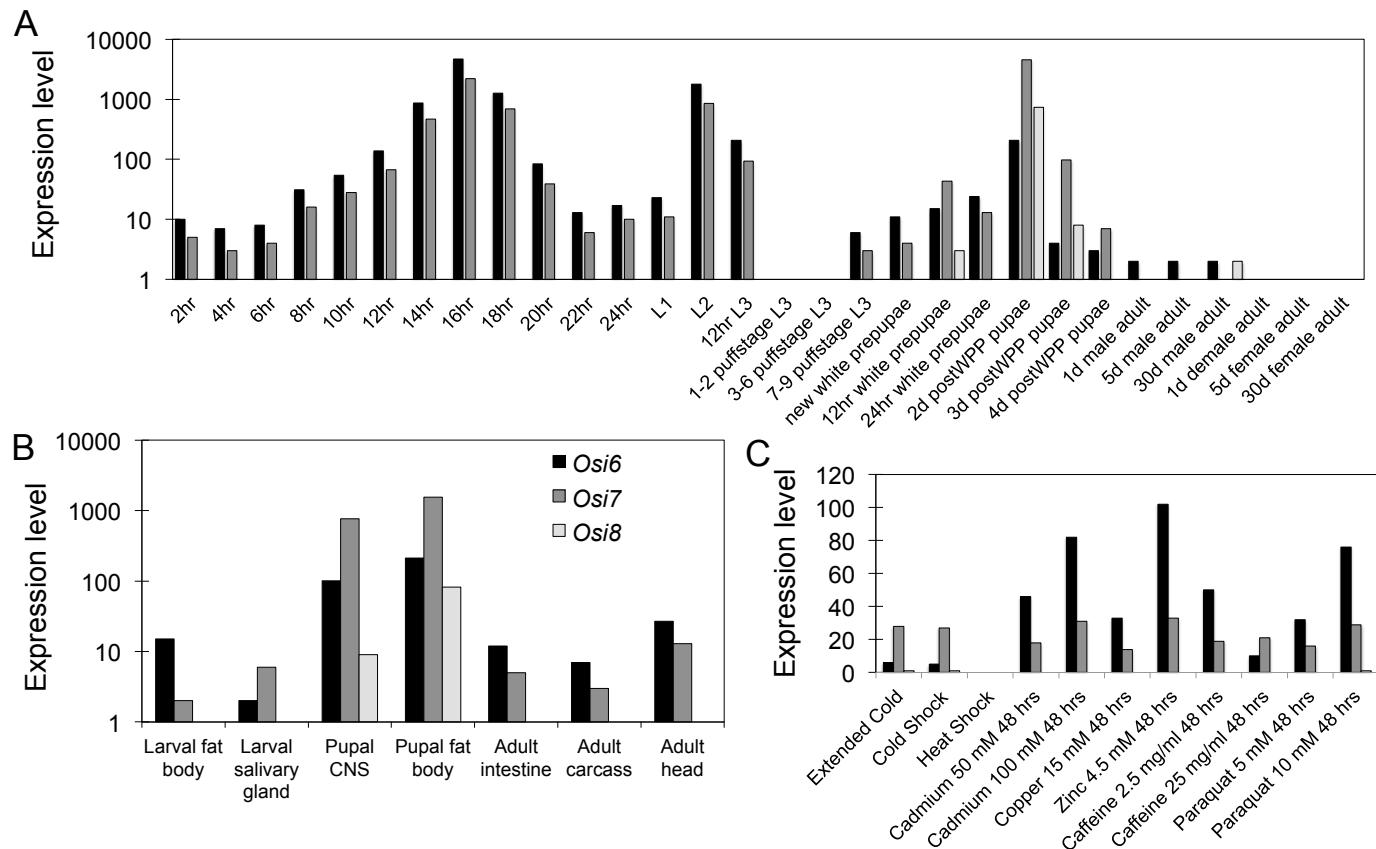


Figure S4

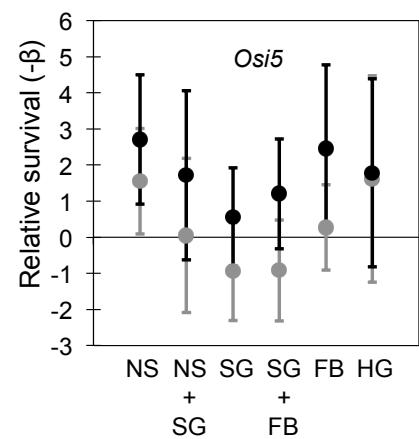


Table S1

Gene	Stock	Genotype	Source
Control	v60000	w ¹¹¹⁸	VDRC
Gasp	v102518	P{KK111875}VIE-260B	VDRC
Obp83cd	v42725	w ¹¹¹⁸ ; P{GD748}v42725	VDRC
Obp83ef	v18814	w ¹¹¹⁸ ; P{GD5605}v18814/CyO	VDRC
Vha14-2	v40807	w ¹¹¹⁸ ; P{GD15762}v40807	VDRC
CG31559	v33967	w ¹¹¹⁸ ; P{GD10297}v33967	VDRC
CG1077	v7552	w ¹¹¹⁸ ; P{GD517}v7552	VDRC
Osi1	v5738	w ¹¹¹⁸ ; P{GD2817}v5738/TM3	VDRC
CG31562	v33970	w ¹¹¹⁸ ; P{GD10298}v33970	VDRC
NPFR	v9605	w ¹¹¹⁸ ; P{GD668}v9605	VDRC
Osi24	v43404	w ¹¹¹⁸ ; P{GD9064}v43404	VDRC
Osi2	v26791	w ¹¹¹⁸ ; P{GD12629}v26791	VDRC
Osi3	v42612	w ¹¹¹⁸ ; P{GD2819}v42612	VDRC
Osi4	v5747	w ¹¹¹⁸ ; P{GD2820}v5747	VDRC
Osi5	v102392	P{KK110481}VIE-260B	VDRC
Osi6	v44545	w ¹¹¹⁸ ; P{GD2821}v44545	VDRC
Osi7	v8475	w ¹¹¹⁸ ; P{GD2822}v8475	VDRC
Osi8	v5753	w ¹¹¹⁸ ; P{GD2823}v5753	VDRC
GAL4	6357	y ¹ w ¹¹¹⁸ ; P{Lsp2-GAL4.H}3	BDSC
GAL4	6870	w ¹¹¹⁸ ; P{Sgs3-GAL4.PD}TP1	BDSC
GAL4	8180	w ¹¹¹⁸ ; P{GawB}DJ717	BDSC
GAL4	30843	w [*] ; P{GawB}c591	BDSC
GAL4	30844	w [*] ; P{GawB}c601 ^{c601}	BDSC
Obp83g	58515	y ¹ w [*] ; Mi{MIC}Obp83g ^{MI12371}	BDSC

Table S2

Species	Strain	N
<i>D. sechellia</i>	14021-0428.08	80
<i>D. sechellia</i>	14021-0428.27	80
<i>D. sechellia</i>	14021-0428.07	137
<i>D. sechellia</i>	14021-0428.03	80
<i>D. sechellia</i>	droSec1	130
<i>D. melanogaster</i>	Oregon-R	510
<i>D. melanogaster</i>	w1118	729
<i>D. melanogaster</i>	Canton-S	588
<i>D. melanogaster</i>	Zhr	160
<i>D. melanogaster</i>	Z30	160
<i>D. melanogaster</i>	dm3	159
<i>D. mauritiana</i>	14021-0421.60	277
<i>D. simulans</i>	14021-0251.195	223
<i>D. simulans</i>	tsimbazaza	210

Table S3

Gene	<i>actin-GAL4</i>			<i>tubulin-P[Switch]</i>	
	UAS-RNAi	Gal4xUAS	Gal4xUAS ^c yo	Gal4xUAS(H)	Gal4xUAS
Gasp	118	100	160	90	120
Obp83cd	160	258	240	146	141
Obp83ef	110	30	135	120	106
Vha14-2	227	210	187	90	100
CG31559	200	200	110	90	90
CG1077	120	200	200	99	109
Osi1	120	170	142	170	148
CG31562	210	200	200	140	130
NPFR	150	260	290	91	90
Osi24	220	198	210	155	128
Osi2	80	190	163	70	90
Osi3	116	190	170	101	94
Osi4	120	258	260	149	128
Osi5	150	180	180	148	137
Osi6	120	30	185	129	119
Osi7	220	103	339	110	110
Osi8	209	230	143	86	70

Table S4

Gene	Forward	Reverse
<i>Osi6</i>	TCTTCCTGGCTCTGGCTGCT	TCTTGCCGCCGAAGAGACCT
<i>Osi7</i>	CGGC GGCGAGAACGACATTA	ACCGAGCACCTTGTCCACGA
<i>Osi8</i>	TGTCCGTTGCCTAAAGGTCA	TCCTTGGCGTCCACACTTC
α Tub84B	TGTCGCGTGTGAAACACTTC	AGCAGGCGTTCCAATCTG
<i>Act42A</i>	GCGTCGGTCAATTCAATCTT	AAGCTGCAACCTCTCGTCA