

1 SUPPORTING INFORMATIONS – Duron, Wilkes and Hurst

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Taxon	Population	<i>n</i>	Infection by <i>Arsenophonus</i>
HYMENOPTERA			
Ichneumonidae			
<i>Phygadeuon fumator</i>	Edwards (Ontario, Canada), 2000	39	-
Pteromalidae			
<i>Nasonia vitripennis</i>	Rochester (New York, USA), 2006	38	3 (8%)
<i>Muscidifurax raptor</i>	St-Anne de Prescott (Ontario, Canada), 2003	44	1 (2%)
	Edwards (Ontario, Canada), 2000	35	-
<i>Spalangia cameroni</i>	Alfred (Ontario, Canada), 2002	44	3 (7%)
	St-Anne de Prescott (Ontario, Canada), 2002	38	-
	St-Anne de Prescott (Ontario, Canada), 2003	32	-
<i>Spalangia nigra</i>	St-Anne de Prescott (Ontario, Canada), 2002	24	-
	Thurso (Québec, Canada), 2002	1	-
	Alfred (Collège) (Ontario, Canada), 2002	3	-
	Alfred (Ontario, Canada), 2002	1	-
	Alfred (Ontario, Canada), 2002	1	-
	Alfred (Ontario, Canada), 2002	1	-
<i>Spalangia nigroaenea</i>	St-Anne de Prescott (Ontario, Canada), 2003	37	-
<i>Trichomalopsis sarcophagae</i>	Wainwright (Alberta, Canada), 1997	3	-
	Lacombe (Alberta, Canada), 1997	1	-
	Ponoka (Alberta, Canada), 1997	1	-
	Westlock (Alberta, Canada), 1997	1	-
	Red Deer (Alberta, Canada), 1997	3	-
	Ryley (Alberta, Canada), 1997	6	-
<i>Trichomalopsis viridescens</i>	Red Deer (Albeta, Canada), 1997	12	-
<i>Urolepis rufipes</i>	Saint Paul (Alberta, Canada), 2000	10	-
	Edwards (Ontario, Canada), 2000	6	-
	Ottawa (Ontario, Canada), 2000	8	-
	Kars (Ontario, Canada), 2000	3	-
	Quyon (Québec, Canada), 2000	6	-
	Luskville (Québec, Canada), 2000	5	-
DIPTERA			
Calliphoridae			
<i>Protocalliphora azurea</i>	Locality not known (Corsica, France), 2003	12	2 (17%)
	Muro (Corsica, France), 2005	19	-
	Pirio (Corsica, France), 2005	83	3 (4%)
	Montpellier (France), 2005	12	-
	Locality not known (Algeria), 2005	20	-

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4 **Table S1** Distribution of *Arsenophonus* among parasitoid wasps and their host flies.

5 Infection was evidenced through PCR amplification of a fragment of *Arsenophonus*

6 *16S rRNA* gene. One parasitoid female per host fly pupa has been checked for

7 infection, minimizing the risk of sampling sibling individuals.

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Gene	Hypothetical product (reference ORF in <i>A. nasoniae</i>)	Primers (5'-3')	Tm	Fragment size	Reference
<i>Arsenophonus</i>					
<i>16S rRNA</i>	Small ribosomal subunit (Genbank M90801)	ArsF ArsR2	GGGTTGTAAAGTACTTTCAGTCGT GTAGCCCTRCTCGTAAGGGCC	52°C	(Duron <i>et al.</i> 2008)
<i>fbaA</i>	Fructose-bisphosphate aldolase class II (ARN_34300)	fbaAf fbaAr	GCYGCYAAAGTTCRTTCTCC CCWGAACCDCCRTGGAAAACAAAA	52°C	659bp This study
<i>ftsK</i>	Cell division protein (DNA translocase) (ARN_16600)	ftsKf ftsKr	GTTGTYATGGTYGATGAATTTGC GCTCTTCATCACYATCAWAACC	52°C	445bp This study
<i>yaeT</i>	Outer membrane protein assembly factor (ARN_26640)	yaeTf yaeTr	GCATACGGTTCAGACGGGTTTG GCCGAAACGCCTTCAGAAAAG	52°C	473bp This study
<i>Arthropods</i>					
<i>COI</i>	cytochrome oxidase subunit I	C1-J-1751a C1-N-2191	GGATCACCTGATATAGCATTCCC CCCGGTAATATAAAATATAAACTTC	50°C	449-451bp (Bogdanowicz <i>et al.</i> 1993)
<i>18S rRNA</i>	Small ribosomal subunit	NSF4/18 NSR399/19	CTGGTTGATYCTGCCAGT TCTCAGGCTCCYTCTCCGG	52°C	418-422bp (Hendriks <i>et al.</i> 1990; Hendriks <i>et al.</i> 1991)

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10 **Table S2** Genes and primer features. For reference to the *A. nasoniae* genome ORFs, see Darby *et al.* (2010).

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Gene	No. strains positive	No. alleles	Variable sites (%)	Nucleotide diversity per site	G + C content (%)	K_a/K_s ratio	Recombination (Sawyer's test P -value)
<i>fbaA</i>	13	8	19.8	0.069	40.5	0.062	No (0.26)
<i>ftsK</i>	14	7	15.0	0.063	46.0	0.092	No (0.93)
<i>yaeT</i>	14	7	10.5	0.039	39.7	0.267	No (0.18)

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13 **Table S3** Amplifiability, diversity and characteristics of the three MLST genes from typing of 14 *Arsenophonus* isolates. Two genes (*ftsK* and
14 *yaeT*) showed positive PCR amplification for all the *Arsenophonus* isolates, but one for 13 isolates (*fbaA* with the *Arsenophonus* strain of the fire
15 bug *Pyrrhocoris apterus* failing to amplify). The DnaSP program (Librado & Rozas 2009) was used to calculate the number of polymorphic site,
16 the %GC content, and the pairwise ratio of nonsynonymous to synonymous substitutions (K_a/K_s). We used Sawyer's test procedure (Sawyer
17 1989) implemented in GENECONV (Sawyer 1999) to perform statistical analysis for intra-genic recombination (10,000 permutations).

Species	number of infected lines (total number of lines tested)			
	F3	F4	F5	F6
<i>Nasonia giraulti</i>	3 (3)	3 (4)	2 (2)	2 (2)
<i>N. longicornis</i>	2 (2)	2 (2)	1 (1)	1 (1)
<i>Muscidifurax raptorellus</i>	5 (5)	5 (5)	3 (4)	3 (3)
<i>Melittobia acasta</i>	5 (5)	3 (3)	3 (3)	2 (3)

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20 **Table S4** Maternal inheritance of *Arsenophonus nasoniae* infection through
21 generations F3-F6 post transfer (F1 and F2 are presented in Table 1). Infection was
22 evidenced through PCR amplification of a fragment of *Arsenophonus nasoniae* *16S*
23 *rRNA* gene.

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