

PROTEIN IDENTITIES- *GRAPHOCEPHALA*
ATROPUNCTATA EXPRESSED SEQUENCE TAGS:
EXPANDING LEAFHOPPER VECTOR BIOLOGY

WAYNE B. HUNTER^{(1)*}, KENT S. SHELBY⁽²⁾, ALEXANDER H. PURCELL⁽³⁾, AND
LAURA E. HUNNICUTT⁽⁴⁾

⁽¹⁾United States Department of Agriculture, Agricultural Research Service, U.S. Horticultural Research Laboratory, Fort Pierce, FL 34945, U.S.A.

⁽²⁾United States Department of Agriculture, Agricultural Research Service, Biological Control of Insects Research Laboratory, Columbia, MO 65203 U.S.A.

⁽³⁾University of California, Division of Insect Biology, Berkeley, CA 94720-3112, U.S.A.

⁽⁴⁾North Carolina State University, Genomic Sciences, 128 Polk Hall, Raleigh, NC 27695 U.S.A.

ABSTRACT: Although *Graphocephala atropunctata* (*Signoret*) (*Hemiptera: Cicadellidae*) is the native blue-green sharpshooter, BGSS, which has been a major vector of Pierce's disease in vineyards in California for nearly a century, only recently has any genomic information become available. Due to the importance of the BGSS as the principal native vector of Pierce's disease, we chose to examine the biology of the BGSS using a genomics approach. A cDNA library was made from adult BGSS, and 8,160 expressed sequence tags, ESTs, were produced. After quality scoring 6,836 sequences underwent assembly which produced a set of 1,915 sequences that putatively represented distinct transcripts. Initial annotation of this dataset identified 44 putative protein sequences were characterized through in silico analyses, and published in the NCBI database (Accession numbers are listed in Table 1). BLASTX analysis identified 10 significant homology matches to heat shock proteins, HSP, which are the focus of this study due to their overall importance and functions in maintaining protein integrity and activity during stressful conditions, such as extreme heat, cold, drought or crowding. A putative full-length small heat shock protein was produced NCBI database accession DQ445538.1. Many other genes of interest which have various functions in leafhopper biology and physiology have also been identified but are not reported herein. The EST sequences reported in this study have been deposited in GenBank's dbEST under accession numbers EH655849–EH662328 and EH662332.

Key Words: EST, DQ445538.1, *Graphocephala*, Heat Shock Proteins, Leafhopper, Sharpshooter, *Xylella*

SHARPSHOOTER leafhoppers are the primary vectors of *Xylella fastidiosa*, the bacterium that causes several destructive plant diseases, including Pierce's disease (PD). The native leafhopper, *Graphocephala atropunctata* (*Signoret*) (*Hemiptera*) blue-green sharpshooter, BGSS, is a major vector of *X. fastidiosa* in vineyards in California and has been spreading PD for nearly a century. Unlike the invasive glassy-winged sharpshooter, GWSS, *Homalodisca vitri-*

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pennis Germar (Takiya et al, 2006) the BGSS is smaller in size and prefers to feed in riparian habitats, thus keeping most infections in grapevine to the bordering plants. The crops grown in the San Joaquin Valley face extremely hot temperatures in the summers, often over 38°C and freezing temperatures during the winter months which creates a highly stressful environment for sharpshooters and other insects. The importance of BGSS as the principal native vector of *X. fastidiosa* in grapes (Almeida et al., 2005a,b) led us to examine the biology of BGSS using a genomics approach to determine how these sharpshooters deal with harsh environmental conditions. The benefits gained from expressed sequence tag, EST, studies have been definitively demonstrated through many studies on insects (*Drosophila*, Honey bee, Aphids, Silk worm, Psyllids) and other organisms. Current production of genomic information on the BGSS is now available (Hunter et al., 2006; 2007). The identification of genes associated with leafhopper biology continues to expand as costs continue to drop and more ESTs are produced from different species. Annotation of these datasets advances the current understanding of leafhopper biological pathways while providing clues to the genetic basis of such processes as insect-pathogen, and insect-plant interactions. The availability of genomic data on BGSS, which is one of three sets of genomic data on different species of sharpshooters (Hunter 2003; Hunter et al., 2005) provides a solid foundation for future studies in functional genomics to advance the development of novel genomics-based management strategies for this and other leafhopper vectors of plant pathogens. Herein we report on the production and annotation of 44 putative proteins from BGSS, and a unique small heat shock protein.

METHODS—Leafhoppers—Adult *G. atropunctata* were obtained from a colony managed by Alexander Purcell at the University of California (Berkeley, CA). Founder BGSS were field-collected from mugwort (*Artemisia douglasiana* L) in Guerneville, CA (Sonoma Co.) and subsequently reared on sweet basil (*Ocimum basilicum* L.) at 25°C (+10°C/–5°C), 14 L: 10 D. First-generation progeny were macerated in RNAlater® RNA Stabilization Reagent (Ambion, Austin, TX) and stored at –40°C prior to shipment to the U.S. Horticultural Research Laboratory, Ft. Pierce, FL.

cDNA Library construction—Approximately 450 adult BGSS of mixed age and genders were used in the construction of an expression library. Whole leafhoppers were ground in liquid nitrogen and total RNA extracted using guanidinium salt-phenol-chloroform procedure as previously described by Strommer and co-workers (1993). Poly(A) + RNA was purified using two rounds of selection on oligo dT magnetic beads according to the manufacturer's instructions (Dynal, Oslo, Norway). cDNA was synthesized using Stratagene's ZAP-cDNA Synthesis Kit (Stratagene, La Jolla, CA, USA). Mass excision of the amplified library was carried out using Ex-Assist helper phage (Stratagene, La Jolla, CA, USA) and bacterial clones containing excised pBluescript SK(+) phagemids were recovered by random colony selection.

Sequencing of clones—pBluescript SK(+) phagemids were isolated from cell cultures grown overnight in 96-deep well plates containing 1.8 mL of LB broth supplemented with 100 µg/mL ampicillin. DNA was extracted using the Qiagen 9600 liquid handling robot and the QIAprep 96 Turbo miniprep kit according to the recommended protocol (QIAGEN Inc., Valencia, CA, USA).

Sequencing reactions were performed using the ABI PRISM® BigDye™ Primer Cycle Sequencing Kit (Applied Biosystems, Foster City, CA, USA). Reactions were prepared in 96-well format using the Biomek2000™ liquid handling robot (Beckman Coulter, Inc., USA). Sequencing reaction products were precipitated with 70% isopropanol, resuspended in 15 µL sterile water and loaded onto an ABI 3730 DNA Analyzer (Applied Biosystems, Foster City, CA, USA).

Sequence analysis—Base calling was performed using TraceTuner™ (Paracel, Pasadena, CA) and Sequencher™ software (Gene Codes, Ann Arbor, MI). Putative sequence identity was determined based on BLAST similarity searches using the NCBI BLAST server (9/12/2008, www.ncbi.nlm.nih.gov) with comparisons made to both non-redundant nucleic acid and protein databases. Matches with an E-value ≤ -10 were considered significant. Protein sequence alignment for the small heat shock protein from *Graphocephala atropunctata*, to: Locust [*Locusta migratoria* gi|85816370], Pink hibiscus mealybug [*Maconellicoccus hirsutus* gi|121543671], Honey bee [*Apis mellifera* gi|11075766], Parasitoid [*Nasonia vitripennis* gi|156553185], Pea aphid [*Acyrtosiphon pisum* gi|193688392], Silverleaf Whitefly [*Bemisia tabaci* gi|198250388], Greenhouse Whitefly [*Trialeurodes vaporariorum* gi|198250396], Beetle [*Tribolium castaneum* gi|91089095], and mosquito [*Aedes aegypti* gi|157135561] (Altschul et al., 1997; Schäffer et al., 2001; Marchler-Bauer and Bryant 2004). Alignment CLUSTAL W2.0.8 multiple sequence alignment tool (<http://www.ebi.ac.uk/Tools/clustalw2/index.html>) (Higgins et al., 1994; Thompson et al., 1997) (Fig. 1).

RESULTS—Sequences—The 5'-single pass sequencing of a cDNA library derived from adult *G. atropunctata* yielded 8,160 ESTs, of which 6,836 were designated as “high quality” (i.e., ≥ 200 bases with a TraceTuner™ score of 20 or better). Translated proteins were analyzed with BLASTP, and ExPASy (Gasteiger et al., 2003). Forty putative proteins identified from BGSS are listed in Table 1. Homologous matches to heat shock proteins, HSP 20, 40, 70, and 90, are shown in Table 2. A putative full-length protein was matched to a small heat shock protein (Table 3). Protein sequence alignment for the small heat shock protein, sHSP, from *G. atropunctata*, to: Locust, Pink hibiscus mealybug, Parasitoid *Nasonia*, Pea aphid, Honey bee, two whitefly species, Beetle *Tribolium*, and *Aedes* Mosquito is shown in Figure 1. A phylogenetic analysis of the *G. atropunctata* sHSP (Fig. 2) produced no clade with other sHSP from the other species.

DISCUSSION—Small heat shock proteins, sHSP, considered α -crystallin proteins, are defined by a conserved sequence of approximately 90 amino acid residues, termed the α -crystallin domain (MacRae, 2000; Taylor and Benjamin, 2005). Functionally, most sHSP display *in vitro* chaperone-like activity, that is, the capacity to interact with other HSP to prevent aggregation and to keep proteins in a folded, competent state (Franck et al., 2004). Small HSP occur in all Kingdoms, but not in all organisms and have been implicated in an astounding variety of processes, such as enhancing cellular stress resistance (Feder and Hofmann, 1999), regulating actin and intermediate filament dynamics (Wieske et al., 2001), inhibiting apoptosis, modulating membrane fluidity (Tsvetkova et al., 2002), and regulating vasorelaxation (Flynn et al., 2003). Amino acid sequence comparisons of the BGSS sHSP with other sHSP showed the common motif of the alpha-crystallin domain, NCBI GenBank database, 9/12/2008 (Altschul et al., 1997, 2005, <http://www.ncbi.nlm.nih.gov/>

Bemisia_tabaci	MQSALRAALLNELVDELTN---PLSALTDQNFQIGILLTDELNSRPRYQHT	47
Trialeurodes	MQSALRAALLNELVDELTN---PLSALTDLNFQIGLMTDELNSRPRYQHT	47
Locusta_migratoria	-----MALVRELFDDLNR---PMY-LFDQNFGLGMLGDDL-LIPIR-----	35
Maconellicoccus_hirsutus	--MSLLPYIVNELVRSYDRYDPPSPLYDQHFGLGLLNDLRYRPAIS--	46
Graphocephala	--MSMVPYIVREMLRDMR----PTLYDQHFGLGLSPANLVDHGL----	39
Nasonia_vitripennis	--MALIPTMFRDWDDLDR---PSRLMDQHFGLMGLTRDEL-----H	37
Acyrtosiphon_pisum	--MSLVPLFFRDWEDFERER-LPRRLLDQHFGLGLHRDDLSNL----TS	43
Apis_mellifera	--MSLIPLLFSDWEDLDR---PHRLLDQNFGLGLYPEQLLSNILDQY	44
Tribolium_castaneum	---MALWLFNDPYDYR----RPSRLHDQHFGLGSLVDPEDLLSP----	36
Aedes_aegypti	--MSLVPMFRDWDDFDSPL-RSSRLLDQHFGLGLRADDLFSS----L	42
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Bemisia_tabaci	ALSPLPLAA----GYVRPWRI SPA--QQSGVSNIHHDKAAFKVNLVDVQQF	90
Trialeurodes	ALSPLPLAA----GYVRPWRI SPA--QQSGVSNIHHDKAAFKVNLVDVQQF	90
Locusta_migratoria	TATVPLLS----GYRPRWRVAT--RHSGTNSIQNTKNDPKVSLVDVQQF	78
Maconellicoccus_hirsutus	AFSTPVLA----GYLRPHRHSHP--ENSGISTIVNQKDKQFKVNLVDVQQF	89
Graphocephala	-LTPMLS----GYLRPWRI LNQ--ADSGLSNIVNDKDNFKVSLVDVQQF	81
Nasonia_vitripennis	TLSPVSPFR----GYFRPWRLLE-QTG-GVSRVQSDKDKFQVI IDVQQF	80
Acyrtosiphon_pisum	ALSSPSLRSA--TYRPPWQGLVN-RQNSGTSLNLFDEKQVQVILVDVQQF	89
Apis_mellifera	ILPNRNLRLRNLPIYYRPPWGLLRKNEGGGTSTVKADKDKFQVILVDVQQF	94
Tribolium_castaneum	IPREFRHYLRSPAGYLRPW-RSLASQD-SGSTVSYDKDKFQACLDVQQF	84
Aedes_aegypti	STRTPSTLLR-SGYRPRWRNTALTRQD-SGSTLNLDDKDKFQI ILVDVQQF	90
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Bemisia_tabaci	QPPEVSVKVVVDGFLVVEAKHEERQDKHGYSRSFTRRYKLPKDINEDAIV	140
Trialeurodes	QPKVSVKVVVDGFLVVEAKHEERQDKHGYSRSFTRRYKLPKDINEDAIV	140
Locusta_migratoria	KPEEINVKVVVDVVI EGKHEERQDEHGFI SRQFTRRYKLPNDVLEAVS	128
Maconellicoccus_hirsutus	KPEEVNWKIVDDYLVVEGKHEERQDKHGYSRSQFTRRYKLPQNVNLEATA	139
Graphocephala	KPEELTVKVVVDCVVVEGKHEERSDEHGFSRQFTRRYRPLNDCDQVALQ	131
Nasonia_vitripennis	GPQEISVKTVDNCI IVEAKHEEKDEHGFI SRQFRRYVLPQEGHDIGNVQ	130
Acyrtosiphon_pisum	GPGEITVKTSEGA IVEGKHEEKQDEHGFI SRQFKRRYLLPKDQVDIEQIV	139
Apis_mellifera	KPDEINVKIVDKCVVVEGKHEEKQDEHGWSRQFTRRYMIPEQCIDIQVT	144
Tribolium_castaneum	KPEEITVKVSDNVVVEGKHEEKQDEHGFI SRHFVRRYMLPKGHVDEKVE	134
Aedes_aegypti	TPPEITVKTDTKYVVEGKHEEKQDEHGFSRHFTRRYMLPSGHDPNDIV	140
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Bemisia_tabaci	SSLSSDGLVTISATVKNQLPSG--ERQIPITQTNQPALKKAKSDAPQENG	188
Trialeurodes	SSLSSDGLVTISATVKNQLPSG--ERQIPITQTNQPALKKAKSDAPQENG	188
Locusta_migratoria	SKLSSDGLVTITAPKKQLSPANSKERVIIQVQTNKPAKLSAPGND--GD	175
Maconellicoccus_hirsutus	SNLSSDGLSITAPKKAENEAK-EISIPVQTNQPAIKQTNKNE--EKS	186
Graphocephala	SSLSSDGLVQLTAPKKSIEDKG--ARPIPTITQTNPAVKAAADGKP---	176
Nasonia_vitripennis	SSLSSDGLVTITAPTLALP-APG-EKI IPIQHTAAPAVKVN-----	169
Acyrtosiphon_pisum	SSLSSDGLITVSPVKKETQ-VTG-ERSVPI IQTGIPAVKAAEAMKNDET	187
Apis_mellifera	SSLSSDGLVNI TAPRKEQPKIQN-ERNITIEQTGPKALKENTEKKEEK-	192
Tribolium_castaneum	SKLSSDGLVTITAPR--VGTEEE-HRSIPIVQTGQPS-KAVEQKKEEKK-	179
Aedes_aegypti	STLSSDGLVTV TAPKKS LAPNP-ERSVPIQQTGQPA-KEQPQSESEVKI	188
	* * : * : : * : * : : * : * : : : *	

(*) denotes similar amino acid, (:) substitution which does not alter protein structure

FIG. 1. Protein sequence alignment for the small heat shock protein from *Graphocephala atropunctata*, to: Locust [*Locusta migratoria* gi|85816370], Pink hibiscus mealybug [*Maconellicoccus hirsutus* gi|121543671], Honey bee [*Apis mellifera* gi|11075766], Parasitoid [*Nasonia vitripennis* gi|156553185], Pea aphid [*Acyrtosiphon pisum* gi|193688392], Silverleaf Whitefly [*Bemisia tabaci* gi|198250388], Greenhouse Whitefly [*Trialeurodes vaporariorum* gi|198250396], Beetle [*Tribolium castaneum* gi|91089095], and mosquito [*Aedes aegypti* gi|157135561]. Alignment using CLUSTAL W2.0.8 multiple sequence alignment tool (<http://www.ebi.ac.uk/Tools/clustalw2/index.html>). (*) denotes similar amino acid, (:) substitution which does not alter protein structure.

blast). The α -crystallin domain is a hallmark of the α -crystallin/small HSP superfamily. The putative α -crystallin domain was present at amino acid positions 64–146. The percentage identity among BGSS to other insect sHSP deduced amino acid sequences varied from 44% to 56% with the highest similarity between *Locusta migratoria* HSP 20.7 and *Maconellicoccus hirsutus*, sHSP, (Table 3) with the lowest similarity to *Rattus norvegicus* (not shown).

TABLE 1. Proteins from *Graphocephala atropunctata*, the blue-green sharpshooter, 44 Putative Protein Sequences, DQ445499–DQ445542. National Center for Biotechnology Information, NCBI. <http://www.ncbi.nlm.nih.gov/sites/entrez>.

Definitions	Clone	Accession Number
file WHGA0016 (similar to CG2210).sqn:	WHGA0016	DQ445499
file WHGA0091 (ribonuclease).sqn:	WHGA0091	DQ445500
file WHGA0096 (cytochrome C oxidase polypeptide):	WHGA0096	DQ445501
file WHGA0097 (s9e ribosomal protein).sqn:	WHGA0097	DQ445502
file WHGA0105 (ubiquitin fusion protein).sqn:	WHGA0105	DQ445503
file WHGA0114 (tropomyosin 1).sqn:	WHGA0114	DQ445504
file WHGA0124 (thioredoxin-like protein).sqn:	WHGA0124	DQ445505
file WHGA0140 (CSF signaling molecule).sqn:	WHGA0140	DQ445506
file WHGA0151 (mitochondrial ATP synthase).sqn:	WHGA0151	DQ445507
file WHGA0169 (cytochrome c reductase).sqn:	WHGA0169	DQ445508
file WHGA0271(LIM protein).sqn:	WHGA0271	DQ445509
file WHGA0283(tumor protein):	WHGA0283	DQ445510
file WHGA0301(oligomycin sensitivity protein):	WHGA0301	DQ445511
file WHGA0310(cytochrome oxidase Va):	WHGA0310	DQ445512
file WHGA0380(ferritin):	WHGA0380	DQ445513
file WHGA0381(calmodulin):	WHGA0381	DQ445514
file WHGA0392(ADP-ATP translocase):	WHGA0392	DQ445515
file WHGA0411(NADH dehydrogenase 1 alpha):	WHGA0411	DQ445516
file WHGA0412(ribosomal protein L23):	WHGA0412	DQ445517
file WHGA0430(Histone3A):	WHGA0430	DQ445518
file WHGA0449(vacuolar ATPase subunit E):	WHGA0449	DQ445519
file WHGA0585(ribosomal protein 4e):	WHGA0585	DQ445520
file WHGA0587(ribosomal protein L37Ae):	WHGA0587	DQ445521
file WHGA0762(ribosomal protein S23e):	WHGA0762	DQ445522
file WHGA0689(elongation factor 1d):	WHGA0689	DQ445523
file WHGA0199 (ribosomal protein 49).sqn:	WHGA0199	DQ445524
file WHGA0225 (V-ATPase).sqn:	WHGA0225	DQ445525
file WHGA0228 (NADH-ubiquinone reductase).sqn:	WHGA0228	DQ445526
file WHGA0230(cytochrome oxidase VIa).sqn:	WHGA0230	DQ445527
file WHGA0257(ribosomal protein L27Ae).sqn:	WHGA0257	DQ445528
file WHGA0270(mitochondrial ATP synthase).sqn:	WHGA0270	DQ445529
file WHGA0783(cytochrome oxidase Vb):	WHGA0783	DQ445530
file WHGA0824(cytochrome c):	WHGA0824	DQ445531
file WHGA0900(tropomyosin):	WHGA0900	DQ445532
file WHGA0927(PPIase):	WHGA0927	DQ445533
file WHGA1072(ribosomal protein L19e):	WHGA1072	DQ445534
file WHGA1215(GABA):	WHGA1215	DQ445535
file WHGA1242(mito. ATP synthase gamma):	WHGA1242	DQ445536
file WHGA1340(mito. porin):	WHGA1340	DQ445537
file WHGA1462(small heat shock protein):	WHGA1462	DQ445538
file WHGA1611(ribosomal protein L18A):	WHGA1611	DQ445539
file WHGA2669(mito. ATP synthase e):	WHGA2669	DQ445540
file WHGA2689(reductase complex QP-C):	WHGA2689	DQ445541
file WHGA3412(ribosomal protein S7e):	WHGA3412	DQ445542



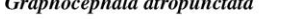


TABLE 2. Heat shock protein homologs to transcripts in *Graphocephala atropunctata*, the blue-green sharpshooter, cDNA library. The full-length cDNA to a small heat shock protein, HSP 20.1, was sequenced and posted in NCBI database accession DQ445538.1. Partial sequences were homologous to HSP 40, HSP 70, and HSP 90.

<i>G. atropunctata</i> Clones	Descriptor	E-value
Contig[1462] 928 bp	gb EAA04497.3 ENSANGP00000018891 Small HSP 20 <i>Anopheles gambiae</i> (Mosquito)	8e ⁻⁴⁰
Contig[1012] 737 bp	gb AAG42838.1 Heat shock protein 70 kDa <i>Leptinotarsa decemlineata</i> (Colorado potato beetle)	6e ⁻²⁸
Contig[1012] 1076 bp	ref XP_623939.1 Heat Shock Protein 90kDa <i>Apis mellifera</i> (Honey bee)	3e ⁻⁸⁴
Contig[1050] 741 bp	dbj BAE44308.1 Heat shock cognate 70 kDa <i>Chilo suppressalis</i> (Striped rice borer)	1e ⁻¹⁰⁴
Contig[1901] 833 bp	gb EAA08691.3 ENSANGP00000012893 HSP cognate 70 kDa, <i>Anopheles gambiae</i> (Mosquito)	6e ⁻⁵⁰
Contig[2381] 857 bp	gb EAA03148.2 ENSANGP00000014454 HSP 40 kDa <i>Anopheles gambiae</i> (Mosquito)	9e ⁻²²
WHGA057-76 560bp	gb AAL27404.1 Heat shock protein 70 kDa <i>Artemia franciscana</i> (Brine shrimp)	2e ⁻⁴⁵
WHGA051-87 578 bp	gb AAO65964.1 Heat shock protein 70 kDa <i>Manduca sexta</i> (Tobacco hornworm)	1e ⁻⁵⁴
WHGA079-33 749 bp	gb AAO21473.1 HSP 70 kDa family <i>Locusta migratoria</i> (Migratory Locust)	2e ⁻⁸²
WHGA008-42 812 bp	dbj BAD74196.1 Heat shock protein 20.1 kDa <i>Bombyx mori</i> (Silkworm)	2e ⁻³⁷

The occurrence of extremely hot temperatures, sometimes over 45°C, in CA and FL produce similar conditions of stress on sharpshooters. The insects must be able to prevent the crosslinking or deformation of proteins to maintain their function and life. Comparative genomics permits us to examine the full

TABLE 3. Alignment of conserved domain for Small Heat Shock Protein, Essential for life, from *Graphocephala atropunctata*, the blue-green sharpshooter. Conserved domain alignments were most similar to *Locusta migratoria* and *Maconellicoccus hirsutus*, sHSP (Expect = 5e⁻⁴⁹), BLAST2, NCBI tools.

Similar alignment 182 aa

DESCRIPTION	SCORE	P	ACCESSION	GI	PROTEIN
<u>Conserved Domain Database hits</u>					
-  <i>Locusta migratoria</i>	569	27	ABC84492	85816366	HSP 20.5 Expect = 5e⁻⁴⁹
-  <i>Graphocephala atropunctata</i>	500	18	AB D98776	90820038	small HSP
-  <i>Maconellicoccus hirsutus</i>	500	18	ABM55532	121543671	small HSP Expect = 5e⁻⁴⁹
-  <i>Apis mellifera</i>	431	18	XP_001...	110750766	Protein lethal(2)essential for life (Protein Efl21)
-  <i>Aedes aegypti</i>	423	18	XP_001...	157135561	lethal(2)essential for life protein, l2efl

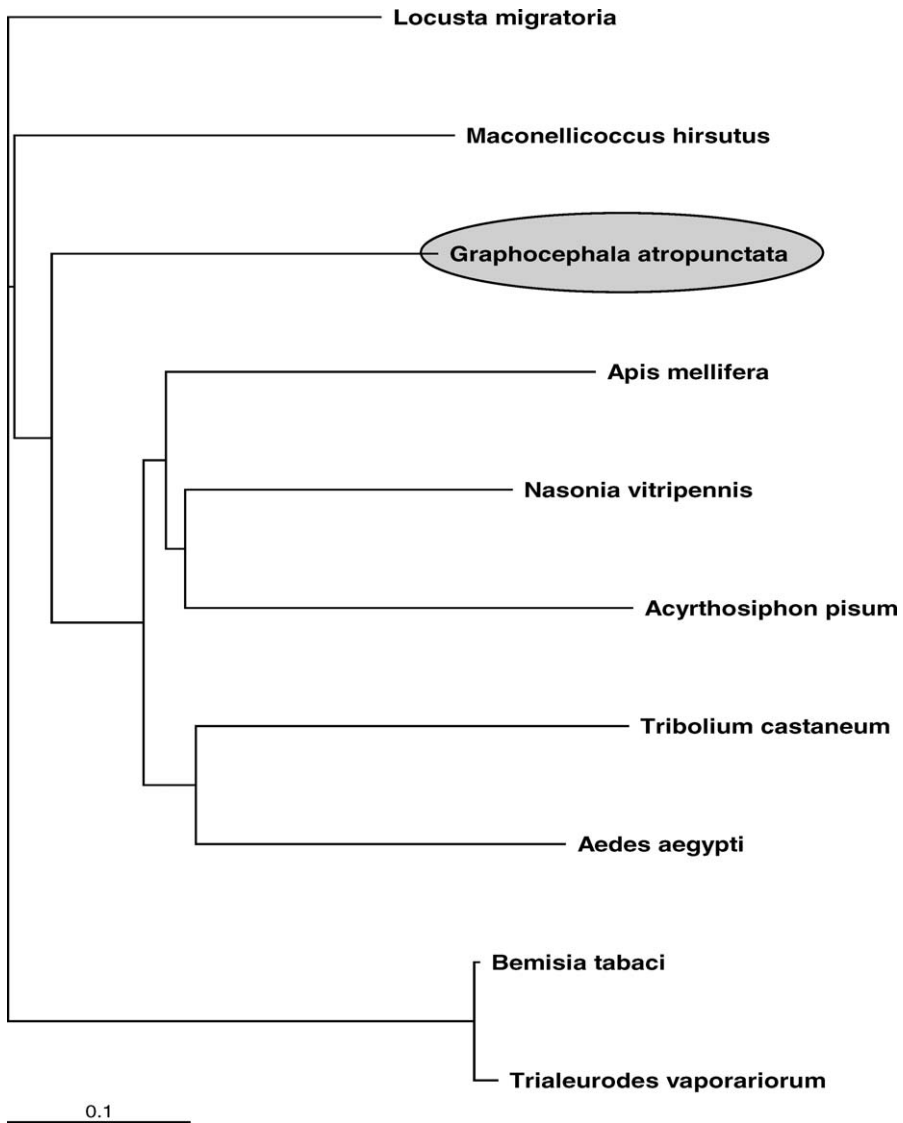


FIG. 2. Phylogenetic tree of the small heat shock protein from *Graphocephala atropunctata*, to: Locust [*Locusta migratoria* gi|85816370], Pink hibiscus mealybug [*Maconellicoccus hirsutus* gi|121543671], Honey bee [*Apis mellifera* gi|11075766], Parasitoid [*Nasonia vitripennis* gi|156553185], Pea aphid [*Acyrthosiphon pisum* gi|193688392], Silverleaf Whitefly [*Bemisia tabaci* gi|198250388], Greenhouse Whitefly [*Trialeurodes vaporariorum* gi|198250396], Beetle [*Tribolium castaneum* gi|91089095], and mosquito [*Aedes aegypti* gi|157135561]. Alignment CLUSTALW2.0.8, NJ, multiple sequence alignment tool.

length cDNAs of HSP 20.5, 20.6, 20.7, 40, 70 and 90 of the migratory locust and other insects which have been cloned and sequenced to make reasonable associations to similar proteins in the BGSS (Singh and Lakhotia, 2000). The functions of HSP are well studied thus providing comparisons between the BGSS and other insects (Ma et al., 2006). We are identifying mRNA transcripts with significant homologies to HSP's in insects like locusts (Qin et al., 2003), to make predictions of HSP's roles in BGSS survival.

The information gained from this study represents the first investigation regarding the transcriptome of *G. atropunctata*, BGSS. The resultant sequence data has produced valuable information on sharpshooter heat shock proteins, and identified many other physiologically important transcripts. The data has been made available to the public to facilitate the use of this information in further studies on sharpshooters. The important role of heat shock proteins to sustain protein integrity and other critical functions make them suitable for further examination as potential genetic targets which may be altered to reduce leafhopper populations. Collectively, these genetic sequences strengthen the foundation needed for further functional genomics studies which will enable the development of more biorational management strategies to reduce losses from the diseases spread by leafhopper pests.

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