



Database tool

WaspBase: a genomic resource for the interactions among parasitic wasps, insect hosts and plants

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Abstract

Insect pests reduce yield and cause economic losses, which are major problems in agriculture. Parasitic wasps are the natural enemies of many agricultural pests and thus have been widely used as biological control agents. Plants, phytophagous insects and parasitic wasps form a tritrophic food chain. Understanding the interactions in this tritrophic system should be helpful for developing parasitic wasps for pest control and deciphering the mechanisms of parasitism. However, the genomic resources for this tritrophic system are not well organized. Here, we describe the WaspBase, a new database that contains 573 transcriptomes of 35 parasitic wasps and the genomes of 12 parasitic wasps, 5 insect hosts and 8 plants. In addition, we identified long non-coding RNA, untranslated regions and 25 widely studied gene families from the genome and transcriptome data of these species. WaspBase provides conventional web services such as Basic Local Alignment Search Tool, search and download, together with several widely used tools such as profile hidden Markov model, Multiple Alignment using Fast Fourier Transform, automated alignment trimming and JBrowse. We also present a collection of active researchers in the field of parasitic wasps, which should be useful for constructing scientific networks in this field.

Database URL: http://www.insect-genome.com/waspbase/

Introduction

Insects are the most widely distributed animal species on earth. Most insects are herbivores that cause huge yield losses when feeding on crops. Insects such as houseflies and mosquitos are vectors of pathogens that cause disease in humans and domesticated animals (1). To combat these insect pests, many methods have been developed, and some of which are used in agriculture. Insecticides are one of the main methods of pest control in agriculture. Unfortunately, overuse of insecticides causes serious environment pollution and food safety problems (2). Therefore, alternative, environment-friendly pest control methods should be developed.

Biological control is an environment-friendly pest control method. Parasitic wasps are well-known biological control agents (3, 4) as they are effective natural enemies of many economically important insect pests. Parasitic wasps are a group of hymenopteran insects that lay eggs in or on the bodies of hosts (5). The wasp larvae feed on the host until pupation and eventually kill the host (6). However, pest control using parasitic wasps has some apparent disadvantages such as wasp development lagging behind pest outbreaks and low-control efficiencies. Understanding the antagonistic interactions between parasitic wasps and their hosts is an important task to improve control efficiencies (7). At present, the genomes of 34 parasitic wasps have been deposited in public databases such as National Center for Biotechnology Information (NCBI). In addition, the genomes of six hosts of these wasps and eight plants that are damaged by these insect hosts are available. Among these species, five parasitic wasps (4,8-11), six insect hosts (12-18) and six plants (19-24) were publicly reported.

Though these data can be retrieved from NCBI, they are not well organized and thus have not been fully explored. Here, we collected the genome and transcriptome data of 34 parasitic wasps, 9 insect hosts and 8 plants from NCBI, i5k workspace@NAL (25) and InsectBase (7). Then, we constructed a database, which we named WaspBase, to serve as an integrated genomic resource for a tritrophic system of wasps, hosts and plants.

Data resources

Genomes

We collected the genome data of 12 parasitic wasps from the NCBI including Ceratosolen solmsi, Copidosoma floridanum, Cotesia vestalis, Diachasma alloeum, Fopius arisanus, Microplitis demolitor, Macrocentrus cingulum, Nasonia giraulti, Niphoparmena longicornis, Nasonia vitripennis, Orussus abietinus and Trichogramma pretiosum (Figure 1) (8, 9, 11). The gene annotation files were

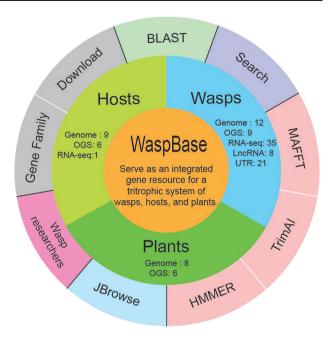


Figure 1. The design of WaspBase. The diagram shows the data and software used in WaspBase.

obtained for nine parasitic wasps including *C. solmsi* (8), *C. floridanum* (10), *D. alloeum*, *F. arisanus* (4), *M. demolitor* (11), *M. cingulum*, *N. vitripennis* (9), *O. abietinus* and *T. pretiosum*. We then focused on these nine parasitic wasps with gene annotation information. There are nine insect hosts for these nine parasitic wasps, of which five have genome data and five have annotated genomes (12, 13). These five insect pests damage eight crops all of which have genome data, but six have annotation information (Figure 2). So, we collected a final genome data of nine parasitic wasps, five insect hosts and six plants (Table 1). The references reporting the interactions between parasitic wasps, insect hosts and plants were given in supplementary table S1.

OGS

The General Feature Format version 3 (Gff3) files containing annotation information were downloaded with the genome data, and the official gene sets (OGSs) were extracted from the genome based on the annotation in the Gff3 file. Then, the nucleotide sequences and protein sequences of annotated genes were produced (Table 2).

Transcriptomes

The raw data of 34 samples of parasitic wasps were downloaded from the NCBI SRA (Sequence Read Archive) database(https://www.ncbi.nlm.nih.gov/sra). We assembled 22 transcriptomes using Trinity and TopHat-Cufflinks with default parameters (26, 27). Together with 21 other

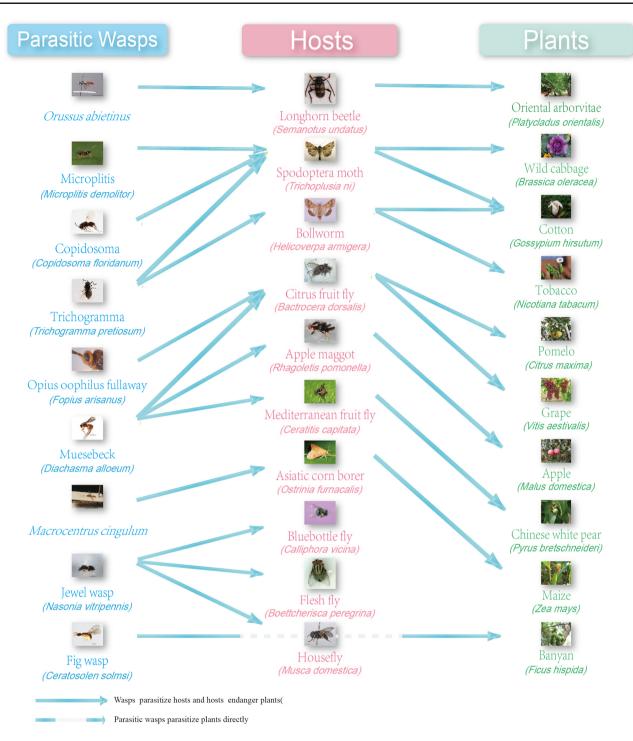


Figure 2. The parasitic wasps, hosts and plants included in the WaspBase. Dashed line: parasitic wasps parasitize plants but not insect hosts. Not dashed line: parasitic wasps parasitize the insect hosts or the insects damage plants.

available transcriptomes, we collected a final transcriptome dataset of 573 RNA-Seq samples from 35 parasitic wasps (Table 3).

IncRNA

Long non-coding RNAs (lncRNAs) are transcribed RNA molecules >200 nucleotides in length that are not protein

coding (28, 29). We predicted lncRNAs of eight parasitic wasps using a previously reported pipeline (30). In total, we predicted 49 607 lncRNAs from eight parasitic wasps.

UTR

We developed a pipeline to predict untranslated regions (UTR) from the transcriptomes and genomes using

	Species name	Accession ID	Source
	Ceratosolen solmsi	GCF_000503995.1	NCBI
	Copidosoma floridanum	GCF_000648655.1	NCBI
	Cotesia vestalis	GCA_001675545.1	NCBI
	Diachasma alloeum	GCF_001412515.1	NCBI
	Fopius arisanus	GCF_000806365.1	NCBI
Wasps	Macrocentrus cingulum	-	InsectBase
	Microplitis demolitor	GCF_000572035.2	NCBI
	Nasonia giraulti	GCA_000004775.1	NCBI
	Nasonia longicornis	GCA_000004795.1	NCBI
	Nasonia vitripennis	GCF_000002325.3	NCBI
	Orussus abietinus	GCF_000612105.1	NCBI
	Trichogramma pretiosum	GCF_000599845.2	NCBI
	Bactrocera dorsalis	GCF_000789215.1	NCBI
	Calliphora vicina	GCA_001017275.1	NCBI
	Ceratitis capitata	GCF_000347755.2	NCBI
Hosts	Helicoverpa armigera	GCF_002156985.1	NCBI
Plants	Helicoverpa zea	GCA_002150865.1	NCBI
	Heliothis virescens	GCA_002382865.1	NCBI
	Manduca sexta	-	InsectBase
	Musca domestica	GCF_000371365.1	NCBI
	Brassica oleracea	GCF_000695525.1	NCBI
	Citrus maxima	GCA_002006925.1	NCBI
	Gossypium hirsutum	GCF_000987745.1	NCBI
	Malus domestica	GCF_000148765.1	NCBI
	Nicotiana tabacum L	GCF_000715135.1	NCBI
	Pyrus x bretschneideri	GCF_000315295.1	NCBI
	Vitis aestivalis	GCA_001562795.1	NCBI
	Zea mays	GCF_000005005.2	NCBI

Table 1. The genome data in the WaspBase

TransDecoder-V5.3.0 (https://github.com/TransDecoder/ TransDecoder), identifying the UTR sequences of 21 parasitic wasps.

Gene families

We used manual annotation by Blastp against known genes (e-value = 10^{-5}), GO annotation and phylogenetic analysis to identify the members of a gene family. We obtained the information of 25 gene families that have been widely studied, including those related to chemoreception, the immune system and detoxification (Figure 3). We also provided a web server for phylogenetic analysis of selected gene members, and we use ClustalW2 (31) to construct a phylogenetic tree by the neighbor-joining clustering method. The bootstrap value was set as 500. The Newick Utilities V1.6 (32) was used to display the phylogenetic tree.

Database construction

Database system implementation

WaspBase was developed on an Apache HTTP (Apache 2.4.25) server in a Linux (RedHat 4.8.2) operating system.

The web pages were written using PHP (PHP 5.6.30), html language, Cascading Style Sheets and JavaScript. All data are stored in the MySQL (MySQL 5.7.17) environment. The Apache server handles queries from web clients through PHP scripts to perform searches.

Search function

WaspBase provides search function using keywords, gene ID, gene names, annotation keywords, KEGG ID, KEGG annotation (33), PFam ID or Pfam annotation (34). Once a gene is searched for, all related gene information was presented in the result webpages. The genes from parasitic wasps, insect hosts and plants were given in the searched results.

Tools module

The tools module contains Basic Local Alignment Search Tool (BLAST) (35), profile hidden Markov model (HMMER), Multiple Alignment using Fast Fourier Transform (MAFFT), automated alignment trimming (TrimAl) and JBrowse (36).

Table 2. The protein and nucleotide dataset in the WaspBase

	Species name	Accession ID	Source
	Ceratosolen solmsi	GCF_000503995.1	NCBI
	Copidosoma floridanum	GCF_000648655.1	NCBI
	Diachasma alloeum	GCF_001412515.1	NCBI
	Fopius arisanus	GCF_000806365.1	NCBI
Wasps	Macrocentrus cingulum	-	InsectBase
	Microplitis demolitor	GCF_000572035.2	NCBI
	Nasonia vitripennis	GCF_000002325.3	NCBI
	Orussus abietinus	GCF_000612105.1	NCBI
	Trichogramma pretiosum	GCF_000599845.2	NCBI
	Bactrocera dorsalis	GCF_000789215.1	NCBI
	Ceratitis capitata	GCF_000347755.2	NCBI
Hosts	Helicoverpa armigera	GCF_002156985.1	NCBI
	Manduca sexta	-	InsectBase
	Musca domestica	GCF_000371365.1	NCBI
	Brassica oleracea	GCF_000695525.1	NCBI
	Gossypium hirsutum	GCF_000987745.1	NCBI
Plants	Malus domestica	GCF_000148765.1	NCBI
	Nicotiana tabacum L	GCF_000715135.1	NCBI
	Pyrus x bretschneideri	GCF_000315295.1	NCBI
	Zea mays	GCF_000005005.2	NCBI

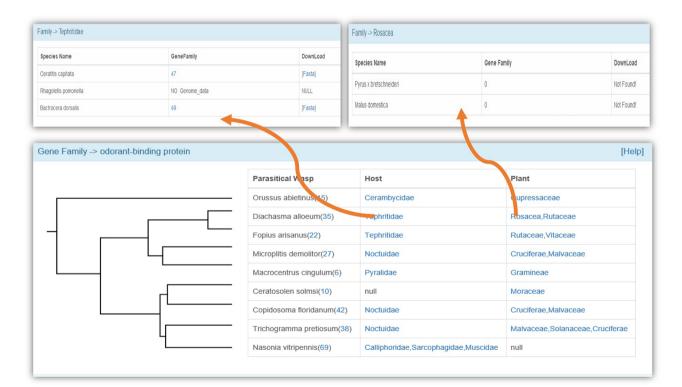


Figure 3. The identified gene families in the WaspBase.

BLAST (35) is provided using the Web-based BLAST server 2.6.0+. The data used for nucleotide BLAST (BLASTN, TBLASTN) searches include 12 insect genomes and 9 insect OGSs. The protein data used for amino acid BLAST (BLASTP, TBLASTX, BLASTX) searches contain nine insect protein sequences. In the BLAST results webpage, users can choose to display top 5 hits, top 10 hits or all hits. The top five BLAST hits are used as default.

Table 3. The transcriptome data in the WaspBase

Species name	Assembly	SRA
Aenasius bambawalei	Trinity	SRR2966926
Anastatus japonicus	Trinity	SRR4034898
Anisopteromalus calandrae	Trinity	SRR2910690,SRR2910691
Asobara tabida	Not assembled	-
Biorhiza pallida	Trinity	ERR1353142,ERR1354102,ERR1354103,ERR1354104,ERR1354105
		ERR1354106,ERR1354107,ERR1354108,ERR1354109,ERR1354110
		ERR1354111,ERR1354112,ERR1354113,ERR1354114,ERR1354115
		ERR1354116,ERR1354117,ERR1354118,ERR1354119,ERR1354354
Ceratosolen solmsi	TopHat-Cufflinks	SRR974922,SRR974923,SRR974924,SRR974925,SRR974926,
		SRR974927,SRR974928,SRR974929
Copidosoma floridanum	TopHat-Cufflinks	SRR1864696,SRR1864697
Cotesia glomerata	Not assembled	-
Cotesia rubecula	Not assembled	-
Cotesia vestalis	Not assembled	-
Diachasma alloeum	TopHat-Cufflinks	SRR2040481,SRR2041626
Diachasmimorpha longicaudata	Not assembled	SRR3336273,SRR3336336,SRR3336337
Diadromus collaris	Not assembled	SRR4294717,SRR1022346
Fopius arisanus	TopHat-Cufflinks	SRR1560649,SRR1560650,SRR1560651,SRR1560653
Leptopilina boulardi	Not assembled	ERR1109367,ERR1109368,ERR1109369,ERR1109370,ERR1109371
E E		ERR1109372,ERR1109373,ERR1109374,ERR1109375,SRR559221,
		SRR559222
Leptopilina clavipes	Trinity	SRR921610
Leptopilina heterotoma	Trinity	SRR559223,SRR559224
Lysiphlebus fabarum	Not assembled	
Macrocentrus cingulum	TopHat-Cufflinks	SRR2968845,SRR2968846
Megastigmus spermotrophus	Trinity	SRR1805073,SRR1805097,SRR1805105,SRR1805115
Microctonus aethiopoides	Not assembled	
Microplitis bicoloratus	Not assembled	_
Microplitis demolitor	TopHat-Cufflinks	SRR955015,SRR955076,SRR955374,SRR955397
Nasonia giraulti	TopHat-Cufflinks	SRR3457435,SRR3457436,SRR3457437,SRR3457438,SRR3457439,
	TopPlat Cullinks	SRR3457457,SRR1566028,SRR1566029,SRR1566030,SRR1566031,
		SRR1566032,SRR1566033,SRR1264518,SRR1264519,SRR1264521,
		SRR1264522,SRR1264523,SRR1264524,SRR1264525,SRR1264526,
		SRR1264527,SRR1264529,SRR1264530,SRR1264531
Nasonia longicornis	Not assembled	-
Nasonia vitripennis	Not assembled	_
Orussus abietinus	TopHat-Cufflinks	ERR1333211,SRR1850925,SRR1850924,SRR921626
Ostrinia furnacalis	Trinity	DRR018822,DRR018823,DRR018824,DRR018825,DRR018826,
		DRR018827,DRR030133,DRR030134,DRR030135,DRR030136,
		DRR030137,DRR030138,DRR030139,DRR030140,DRR030141,
		DRR030142,SRR1032037,SRR1032038,SRR1226611,SRR1265986,
		SRR1560699,SRR1560709,SRR1560711,SRR1565323,SRR1640337,
		SRR1640339,SRR1640341,SRR3189772,SRR3204354,SRR3204356,
		SRR3204357,SRR3374123,SRR3374124,SRR3374125
Psyttalia concolor	Trinity	SRR1593901,SRR1593902
Psyttalia lounsburyi	Trinity	SRR1593906,SRR1593907,SRR1593908
Pteromalus puparum	Not assembled	-
Spalangia endius	Trinity	- SRR2954670,SRR2954673,SRR2954678,SRR2954681,SRR2954683,
spananzia cnains	1111111	SRR2954670, SRR2954675, SRR2954676, SRR2954676, SRR2954686, SRR2954688, SRR2954692, SRR2954704, SRR2954706,
		SRR2954666,SRR2954666,SRR2954666,SRR29546672,SRR2954704,SRR2954706, SRR2954708,SRR2954710,SRR1038395
Telenomus podisi	Trinity	SRR2934/08,SRR2934/10,SRR1038395 SRR1274857,SRR1274858
Telenomus poaisi Trichogramma chilonis	Trinity	SRR12/483/,SRR12/4838 SRR3756972,SRR3756974,SRR3756975,SRR3756979
Trichogramma chilonis Trichogramma pretiosum	TopHat-Cufflinks	SRR3/369/2,SRR3/369/4,SRR3/369/3,SRR3/369/9 SRR1826957,SRR1826958
Venturia canescens		SRR1826937,SRR1826938 ERR791800
venuntu cunescens	Trinity	LIXIX///1000

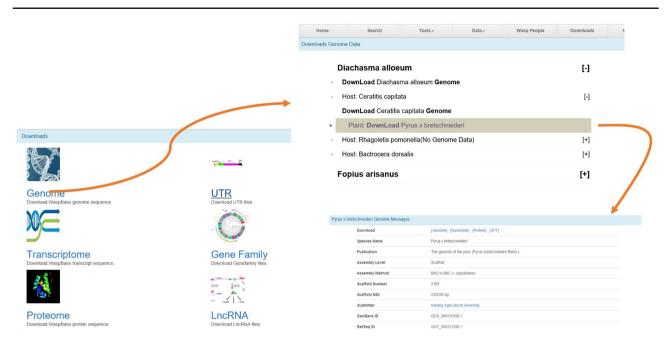


Figure 4. The Download page of WaspBase. The genomes, transcriptomes and OGSs of parasitic wasps and insect hosts are provided together for the convenience of download.

User can also adjust other parameters such as similarity percentage and BLAST score. Links of the BLAST hits were given to directly connect to NCBI for full annotation information. All sequence can be downloaded.

Multiple sequence alignment (MSA) is important for evolutionary analyses. MAFFT (37) is a widely used program for MSA analysis because of its high performance. WaspBase provides a web server of MAFFT and uses TrimAl to trim the aligned sequences (38). To use MAFFT web server, users need to input the sequences in FASTA format with either the default parameters or the customized parameters. To use TrimAl, users need to input the aligned sequences at the TrimAl webpage. The trimmed sequences are showed at TrimAl result webpage. If the number of sequence is more than four, a phylogenetic tree can be constructed using the abovementioned method.

A web server of HMMER is provided to search sequence homologs and to make sequence alignments. It uses probabilistic models called profile hidden Markov models (profile HMMs) (39). To use HMMER, users input the protein sequences at the HMMER webpage. After running the HMMER, the protein sequences are used to search against the Pfam database and the results of protein domain information will be showed at the HMMER result webpage.

Genome visualization

JBrowse is a well-known browser that displays genome annotations by integrating the databases and interactive web pages (36). We used JBrowse in WaspBase to provide interactive views of annotations along with the genome scaffolds. The genome data and the Gff3 files required for JBrowse are stored in a MySQL database using preparerefseqs.pl, flatfile-to-json.pl, add-bam-track.pl and addtrack-json.pl provided by BioPerl. In WaspBase, JBrowse visualizes the annotations and transcriptomes as tracks on the browser for Coding Sequence and coverage of the transcriptome reads. Pop-up balloons in the gene model track display links to gene sequences of interest.

Wasp researchers

To construct a scientific network in the field of parasitic wasp research, we performed reference mining of parasitic wasp studies, which yielded 189 references. Based on publications in the last 5 years, we collected a list of active researchers studying parasitic wasps.

Download

All data can be downloaded, including genomes, transcriptomes, UTR, Gene families and lncRNA. For the convenience of downloading, the gene data of parasitic wasps, insect pests and plants are provided for download at the same webpage (Figure 4).

Conclusions

We constructed WaspBase for parasitic wasps and their corresponding insect hosts and plants. WaspBase provides conventional functions of search, download, domain analysis and phylogenetic analysis, JBrowse display of annotations and other functions described herein. In addition to genomes and transcriptomes, WaspBase also provides lncRNA, UTR and gene family information. A typical feature of WaspBase is that we integrated the gene information of parasitic wasps, their insect hosts and plants targeted by insect pests. Thus, gene data of the tritrophic system in food chains (parasitic wasp–insect pest–plant) were analyzed together, which should be useful for studying cross-species regulation in parasitism and convergent evolution analysis among wasps, hosts and plants.

Future plan

- As the cost of sequencing has been significantly reduced in recent years, the genomes of an increasing number of parasitic wasps will be sequenced. We plan to update WaspBase periodically to keep the database up-to-date.
- 2. Genome annotation is still a time-consuming task and significantly lags behind genome sequencing. We noticed that a number of parasitic wasp genomes are not annotated at present though their genome sequences have been uploaded in the NCBI genome database. We will annotate these genomes using OMIGA (Optimized Maker-Based Insect Genome Annotation) (40), a genome annotation pipeline that we developed.
- 3. It is important to understand cross-species regulation mechanisms and convergent evolution in parasitism. To this end, we will carry out a systematic analysis of more gene families from the OGSs of 'wasps-insects-plants', which should be useful to improve control efficiencies in biological control.

Supplementary data

Supplementary data are available at Database Online.

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Conflict of interest. None declared.

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