Data Article

Transcriptome data of control and *Ascosphaera apis* infected *Apis mellifera ligustica* larval guts

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**Abstract**

*Ascosphaera apis* is an obligate fungal pathogen of honeybee larvae that leads to chalkbrood, which causes heavy losses for the apiculture in China and many other countries. In this article, guts of 4-, 5-, 6-day-old *Apis mellifera ligustica* larvae challenged by *A. apis* (AmT1, AmT2, AmT3) and normal 4-day-old larval guts (AmCK) were sequenced using next-generation sequencing technology. On average, 29,196,197, 28,690,943, 29,779,715 and 30,496,725 raw reads were yielded from these four groups; an average of 29,540,895 clean reads were obtained after quality control. In addition, the mapping ratio of clean reads in treatment and control groups to the *Apis mellifera* genome were over 97.16%. For more insight please see “Uncovering the immune responses of *Apis mellifera ligustica* larval gut to *Ascosphaera apis* infection utilizing transcriptome sequencing” [1]. The raw data were submitted to the National Centre for Biotechnology Information (NCBI) Sequence Read Archive (SRA) database under accession numbers: SRR4084091, SRR4084092, SRR4084095, SRR4084096, SRR4084097, SRR4084098, SRR4084099, SRR4084100, SRR4084101, SRR4084102, SRR4084093, SRR4084094.

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Data description

On average, 29,196,197, 28,690,943, 29,779,715 and 30,496,725 raw reads were respectively generated from AmT1, AmT2, AmT3 and AmCK (Table 1). After strict quality control, an average of 29,540,895 clean reads were obtained, and the ratio of clean reads were over 97.16% in each group (Table 1). Additionally, the mapping ratio of clean reads in each group to the reference genome of A. mellifera were ranged from 86.25% to 89.45% (Table 2). The raw data were deposited in the Sequence Read Archive (SRA) database (http://www.ncbi.nlm.nih.gov/sra/) under accession numbers: SRR4084091, SRR4084092, SRR4084095, SRR4084096, SRR4084097, SRR4084098, SRR4084099, SRR4084100, SRR4084101, SRR4084102, SRR4084093, SRR4084094.
2. Experimental design, materials, and methods

2.1. Honeybee larval gut materials

*A. m. ligustica* larvae were reared following the previously described method [2]. The diet was mixed and frozen in smaller aliquots and was pre-heated to 34°C before use. Two-day-old larvae were taken from the combs with a Chinese grafting tool and carefully transferred to the diets (10 mL). The larvae were fed once a day with 20 mL (3-day-old), 30 mL (4-day-old), 40 mL (5-day-old) and 50 mL (6-day-old) diets adding up to 150 mL diet in total.

Based on the method described by Jensen et al. [3], a fresh chalkbrood mummy was sterilized using 10% sodium hypochlorite for 10 min and then rinsed in sterile distilled water for 2 min; subsequently, the mummy was cut into smaller pieces and cultured at 25°C on plates of Potato dextrose agar (PDA) medium, and at 7 days after culturing, fresh spores of *A. apis* were purified and used to feed 3-day-old larvae (treatment group) at a final concentration of 10^7 spore/mL. The larvae consumed all diet were used for further study, whereas the larvae cannot consumed all diets were discarded. Three-day-old larvae in control groups were fed with an artificial diet without *A. apis* spores. Culture plates (NEXT, China) were incubated at 95% RH and 33°C according to the method described by Aronstein et al. [4].

The honeybee larval gut is the main site *A. apis* parasitizes and larvae usually die before the prepupa stage. Twenty-one guts of 4-, 5- or 6-day-old *A. apis*-infected groups (AmT1, AmT2, and AmT3), 21 guts of 4-day-old honeybee larvae from control group (AmCK) were harvested, immediately frozen in liquid nitrogen and stored at −80°C.

| Table 1 | Overview of deep sequencing and data quality control. |
|---------|------------------------------------------------------|
| Sample  | Raw reads num | Clean reads num | Adapter sequence | Low quality | Poly A | N   |
| AmT1-1  | 28,774,864    | 28,407,618 (98.72%) | 106,791 | 75,473 | 26 | 1333 |
| AmT1-2  | 29,564,924    | 29,262,394 (98.98%) | 90,115 | 59,827 | 12 | 1311 |
| AmT1-3  | 30,217,832    | 29,918,578 (99.01%) | 84,510 | 65,005 | 13 | 99 |
| AmT2-1  | 31,786,112    | 31,429,194 (98.88%) | 97,180 | 81,265 | 14 | 0 |
| AmT2-2  | 26,715,638    | 26,405,020 (98.84%) | 90,573 | 63,519 | 12 | 1205 |
| AmT2-3  | 28,544,942    | 28,238,616 (98.93%) | 81,579 | 71,478 | 7 | 98 |
| AmT3-1  | 31,163,300    | 30,860,010 (99.03%) | 85,022 | 65,121 | 66 | 1436 |
| AmT3-2  | 29,534,074    | 28,720,324 (97.24%) | 100,095 | 306,303 | 31 | 446 |
| AmT3-3  | 30,629,132    | 29,758,812 (97.16%) | 114,989 | 319,668 | 25 | 478 |
| AmCK-1  | 31,148,146    | 30,802,898 (98.89%) | 88,842 | 83,735 | 27 | 20 |
| AmCK-2  | 31,152,632    | 30,802,898 (98.88%) | 101,983 | 72,783 | 20 | 81 |
| AmCK-3  | 30,237,796    | 29,884,378 (98.83%) | 109,717 | 65,649 | 5 | 1338 |

| Table 2 | Summary of mapping clean reads to the reference genome of *Apis mellifera*. |
|---------|--------------------------------------------------------------------------|
| Sample  | Total reads num | Unmapped reads num | Unique mapped reads num | Multiple mapped reads num | Mapping ratio |
| AmT1-1  | 27,942,952      | 3,369,197           | 24,306,993              | 266,762                  | 87.94%        |
| AmT1-2  | 29,045,230      | 3,180,318           | 25,597,778              | 267,134                  | 89.05%        |
| AmT1-3  | 29,759,616      | 3,202,462           | 26,288,338              | 268,816                  | 89.24%        |
| AmT2-1  | 30,908,790      | 3,545,971           | 27,066,899              | 295,920                  | 88.53%        |
| AmT2-2  | 26,025,206      | 3,098,226           | 22,694,714              | 232,266                  | 88.10%        |
| AmT2-3  | 27,965,688      | 2,996,833           | 24,701,137              | 267,718                  | 89.28%        |
| AmT3-1  | 29,849,508      | 4,091,689           | 25,410,587              | 347,232                  | 86.29%        |
| AmT3-2  | 28,575,268      | 3,801,677           | 24,469,781              | 303,810                  | 86.70%        |
| AmT3-3  | 29,512,734      | 4,059,196           | 25,166,090              | 287,448                  | 86.25%        |
| AmCK-1  | 30,540,156      | 3,381,653           | 26,848,499              | 309,654                  | 88.93%        |
| AmCK-2  | 30,544,824      | 3,221,171           | 26,983,693              | 339,670                  | 89.45%        |
| AmCK-3  | 29,636,402      | 3,240,646           | 26,010,060              | 385,696                  | 89.07%        |
2.2. Total RNA isolation, cDNA library preparation and illumina sequencing

Total RNAs were extracted from each sample (pool of seven larval guts) using a TRIzol Kit (Promega, USA) according to the manufacturer’s instructions. RNA quality was checked with a 2100 Bioanalyzer (Agilent Technologies, USA) and RNase-free agarose gel electrophoresis. The total RNA concentration was detected with a 2100 Bioanalyzer. RNA samples were stored at –80 °C for later library construction and next-generation sequencing. Experiments were conducted using three replicas for each sample from both A. apis-treated groups and control group. Totally, 12 RNA libraries were constructed, representing larval samples from A. apis-infected groups and control group. The libraries were as follows: AmT1-1, AmT1-2 and AmT1-3 were replicate libraries for 4-day-old larvae from the A. apis-treated group, AmT2-1, AmT2-2 and AmT2-3 were replicate libraries for 5-day-old larvae from the A. apis-treated group, AmT3-1, AmT3-2 and AmT3-3 were replicate libraries for 6-day-old larvae from the A. apis-treated group, and AmCK-1, AmCK-2 and AmCK-3 were replicate libraries for 4-day-old larvae from the control group.

Oligo (dTs) were used to isolate poly (A) mRNA, which was then fragmented followed by cDNA synthesis using random hexamers. Next, second-strand cDNAs were synthesized using RNase H and DNA polymerase I. The double-stranded cDNAs were purified using the QiaQuick PCR extraction kit (Qiagen, Germany). The required fragments were purified by agarose gel electrophoresis and then enriched by PCR amplification. Finally, the amplified fragments were sequenced on the Illumina HiSeq™ 2500 platform (GeneDenovo Co., China) using 125 bp pair-end strategy. The raw data produced in this research have been deposited in the NCBI SRA Database under accession numbers: SRR4084091, SRR4084092, SRR4084095, SRR4084096, SRR4084097, SRR4084098, SRR4084099, SRR4084100, SRR4084101, SRR4084102, SRR4084093, SRR4084094.

2.3. Quality control and mapping of RNA-seq data

Firstly, the original sequencing image data were transferred into sequence data via base calling, which is defined as raw data or raw reads stored in the fastq format. Secondly, raw reads of all 12 samples were pre-processed by removing adaptor sequence and reads with more than 5% unknown nucleotides; low-quality reads, defined as reads where the percentage of low-quality bases of quality (Q) value ≤ 5 was more than 50% in a read, were also removed. Finally, the clean reads were aligned to the A. mellifera genome assembly Amel_4.5 (http://www.ncbi.nlm.nih.gov/ genome/48?genomeAssemblyId=22683) using SOAP aligner/soap2 with the threshold that no more than two mismatches were permitted in the alignment.

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Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.dib.2020.105264.
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