



Draft genome sequence of the Algerian bee *Apis mellifera intermissa*



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ABSTRACT

Apis mellifera intermissa is the native honeybee subspecies of Algeria. *A. m. intermissa* occurs in Tunisia, Algeria and Morocco, between the Atlas and the Mediterranean and Atlantic coasts. This bee is very important due to its high ability to adapt to great variations in climatic conditions and due to its preferable cleaning behavior. Here we report the draft genome sequence of this honey bee, its Whole Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession JSUV00000000. The 240-Mb genome is being annotated and analyzed. Comparison with the genome of other *Apis mellifera* sub-species promises to yield insights into the evolution of adaptations to high temperature and resistance to Varroa parasite infestation.

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Specifications

Organism	<i>Apis mellifera intermissa</i> (HAL-Blida)
Sex	Male
Sequencer or array type	Illumina
Data format	Analyzed
Experimental factors	N/A
Experimental features	Genome assembly from Illumina whole genome sequencing
Consent	Allowed for reuse citing original author
Sample source location	Algeria

Apis mellifera intermissa, the native honeybee subspecies of Algeria and most of North Africa, is distinctly darker [13] with some times light illumination on the tergites. This honey bee is characterized by a small size, nervousness, aggressive defense behavior and an abundant use of propolis [12]. *A. m. intermissa* is prone to swarming [7], produces many broods and can build up to one hundred queen cells [6]. *A. m.*

intermissa has the high ability to adapt to great variations in climatic conditions [4].

Ritter [11] described the tolerance of *A. m. intermissa* to Varroa mite, *Varroa destructor*, and some studies showed that *A. m. intermissa* is able of self-defense against this mite [5]. Moreover it has a higher level of hygienic behavior compared with several other subspecies [8] and it is characterized by a good cleaning ability [2].

Some pathogens and viruses had been detected, at a low level, in *A. m. intermissa* colonies like the American foulbrood parasite *Paenibacillus larvae*; *Nosema* [1,3] and the deformed wing virus [9]. Such abilities of adaptation to variable climatic conditions, hygienic behavior, Varroa mite tolerance, low infestation with American foulbrood, *Nosema*, and DWV virus makes *A. m. intermissa* a honey bee with very high potential for selection base in breeding programs for honeybees' resistant to diseases and pathogens and more adaptive to the global climatic changes.

Therefore pure lines samples of *A. m. intermissa* collected from Blida (36°31'N 2°58'E) [10] and confirmed by Adjlane and Haddad [2] for their preferable cleaning behavior were selected as a reference sample to be sequenced for its full genome, which will be an important source of information for honeybee research community worldwide.

Here, we report the draft genome sequence of *A. m. intermissa*, which was obtained through sequencing using the Illumina GAIIX instrument. The raw data obtained were trimmed at either end based on the quality score analysis performed using SeqQC tool. Poor or bad quality bases, probably originating from sequencing mis-calls, were trimmed off

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before subjecting it to the assembly software. After the primary analysis of the assembly described below, based on contig sizes, genome representation and its functional elements, the output will be taken up for further analysis.

Raw data QC

The Illumina GAIIIX paired end raw reads were quality checked using Genotypic Pvt. Ltd., proprietary tool SeqQC. Illumina library has uniform read length of 100.

Raw data processing

Illumina paired-end raw reads were processed by perl script for removal of adapters, lowquality bases, and b-blocks trimming towards 3'-end of the read. Coverage is calculated on the assumption of the 240 Mb genome size of *A. m. intermissa* as shown in Supplement.

De-novo assembly of sequenced genome data

Short reads paired-end read data, 85.5 million read pairs were assembled with SOAP *denovo* assembler [14]. Various hash-lengths were tested for *denovo* assembly, ranging from 45 to 65 km. The result shows that 59 hash-length has assembled the best assembly in terms of total contig length, number of contigs generated, and N50 value. The *denovo* assembly generated 2330251 contigs. SOAP *denovo* was used with default parameters. Scaffolding was completed using the software SSPACE 2.0 scaffolder using paired-end data [15]. Scaffolding resulted in 522,976 scaffolds. Although SOAP *denovo* uses paired reads to resolve repeats and polymorphism, it does not perform “gapped” scaffolding: if there is a coverage gap between a read pair, this pair is not utilized. To leverage such pairs and to make better use of long insert libraries, scaffolding is carried out. SSPACE scaffolder is used with these parameters: minimum number of overlapping bases with the seed is 45, minimum overlap required between contigs to merge adjacent contigs in a scaffold is 50, minimum read pairs to compute scaffold is 5, contig extension is switched on, minimum number of reads needed to call a base during an extension is 20, and maximum number of allowed gaps during mapping with Bowtie Langmead et al. [16] is 1. Scaffolds do consist of uncalled bases (Ns). To fill these inter-scaffold Ns with nucleotides, gap closure is done using Gap Closer tool later. With the help of SSPACE scaffolding using Illumina library, the assembly showed significant improvement in N50 value and significant reduction in number of contigs. The draft genome (scaffolds) resulted 522,976 sequences which gives a total genome length of 243 Mb as shown in Supplement.

The draft genome information reported here provides opportunity for further research into the mechanism involved in adaptations that allow the organisms to thrive in high temperature environments and how these bees have good immunity and are resistant to infestation of Varroa parasite.

Summary

Apis m. intermissa is the native honeybee subspecies of Algeria. *A. m. intermissa* occurs in Tunisia, Algeria and Morocco, between the Atlas and the Mediterranean and Atlantic coasts [12], in an area of more than 2500 km long. *A.m. intermissa* indicates the position through this bee races between tropical Africa and European breeds [17]. This bee is very aggressive, nervous and produces many broods with many queen cells [6]. *A. m. intermissa* is prone to swarming, shows a defensive behavior and an abundant use of propolis [12]. Here we report the draft genome sequence of this honey bee. This Whole

Genome Shotgun project has been deposited at DDBJ/EMBL/GenBank under the accession JSUV00000000 (<http://www.ncbi.nlm.nih.gov/nuccore/JSUV00000000>). The version described in this paper is version JSUV01000000. The 240-Mb genome is being annotated and analyzed. Comparison with the genome of other *Apis m. intermissa* sub-species promise to yield insights into the evolution of adaptations to high temperature and resistance to Varroa parasite infestation.

Competing interests

The authors have declared that no competing interest exists.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.gdata.2015.01.011>.

References

- [1] N. Adjlane, S. Kechih, N. Haddad, Comparative study between techniques for the diagnosis of American foulbrood (*Paenibacillus larvae*) in honeybee colony. *J. Anim. Vet. Adv.* 13 (16) (2014) 970–973.
- [2] N. Adjlane, N. Haddad, The first data on hygienic behavior of *Apis mellifera intermissa* in Algeria. *J. Biol. Earth Sci.* 4 (1) (2014) 1–5.
- [3] N. Adjlane, T. El-Ounass, N. Haddad, Population Dynamics of *Varroa destructor* in Colonies of *Apis mellifera intermissa* in Algeria. 10th COLOSS Conference, 6–8th September 2014, Murcia/Spain, 2014.
- [4] C. Barour, A. Tahar, M. Baylac, Forewing shape variation in Algerian honey bee populations of *Apis mellifera intermissa* (Buttel-Reepen, 1906) (Hymenoptera: Apidae): a landmark-based geometric morphometrics analysis. *Afr. Entomol.* 19 (2011) 11–22.
- [5] O. Boecking, W. Ritter, Grooming and hygienic behaviour of *Apis mellifera intermissa* in Tunisia against *Varroa jacobsoni*. *J. Apic. Res.* 32 (1993) 127–134.
- [6] H. Clément, E. Bruneau, J.M. Barbançon, P. Bonnaffé, R. Domérgo, G. Fert, Y. Le Conte, J. Ratia, C. Reeb, B. Vaissière, Le traité Rustica de l'apiculture. *Traité Rustica*, Paris, 2002. 528.
- [7] C. Gadbin, J.M. Cornuet, J. Fresnaye, Approche biométrique de la variété locale d'*Apis mellifera* L. dans le sud tchadien. *Apidologie* 10 (1979) 137–148.
- [8] J.A. Kefuss, Honey bee hygienic behavior: France, Tunisia and Chile. *Apidologie* 26 (1995) 24–26.
- [9] W. Loucif-Ayad, A. Chefrour, M. Algharibeh, N. Haddad, First detection of deformed wing virus of honeybees in Algeria. *Phytoparasitica* 41 (4) (2013) 445–447.
- [10] W. Loucif-Ayad, M. Achou, H. Legout, M. Alburaki, L. Garnery, Genetic assessment of Algerian honeybee populations by microsatellite markers. *Apidologie* (2014) <http://dx.doi.org/10.1007/s13592-014-0331-0>.
- [11] W. Ritter, Development of Varroa mite population in treated and untreated colonies in Tunisia. *Apidologie* 21 (1990) 368–370.
- [12] F. Ruttner, Biogeography and Taxonomy of Honey Bees. Springer, Berlin, 1988.
- [13] T. Shaibi, S. Fuchs, R.F.A. Moritz, Morphological studies of honeybees (*Apis mellifera*) from Libya. *Apidologie* 40 (2) (2009) 97–105.
- [14] R. Luo, B. Liu, Y. Xie, Z. Li, W. Huang, J. Yuan, G. He, Y. Chen, Q. Pan, Y. Liu, J. Tang, G. Wu, H. Zhang, Y. Shi, Y. Liu, C. Yu, B. Wang, Y. Lu, C. Han, D.W. Cheung, S.M. Yiu, S. Peng, Z. Xiaoqian, G. Liu, X. Liao, Y. Li, H. Yang, J. Wang, T.W. Lam, J. Wang, SOAPdenovo2: an empirically improved memory-efficient short-read de novo assembler. *Giga. Sci.* 1 (1) (2012) 18.
- [15] M. Boetzer, W. Pirovano, SSPACE-LongRead: scaffolding bacterial draft genomes using long read sequence information. *BMC Bioinf.* 15 (2014) 211. <http://dx.doi.org/10.1186/1471-2105-15-211>.
- [16] B. Langmead, C. Trapnell, M. Pop, S.L. Salzberg, Ultrafast and memory-efficient alignment of short DNA sequences to the human genome. *Genome Biol.* 10 (2009) R25.
- [17] Ch. Peyvel, L'espèce *Apis mellifera* : les grandes races géographiques. *Bull. Tech. Apic.* 21 (1994) 129–138.