

# Report

## **Genetic characterization of the honeybee (*Apis mellifera* L.) populations in Estonia**

2023

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## Introduction

The honeybee *Apis mellifera* Linnaeus, 1758 has a wide distribution across various climatic conditions. The species is subdivided into more than 30 subspecies recognized by morphological and genetic methods, ten of which are native to Europe [1]. The subspecies or geographic races are restricted in their distribution to specific geographic areas. Moreover, subspecies encompass a diverse array of ecotypes and breeding lines, demonstrating a notable diversity and their capacity to adapt to regional climate conditions, local vegetation, as well as resist pests and pathogens [2, 3]. Estonia is within the distribution area of the European dark honeybee (*A. m. mellifera*), although other subspecies have been imported by humans over the years. Despite their mating independence, both managed and feral honeybee colonies are significantly impacted by human influence.

The use of morphological, genetic and ecological data has allowed the identification of major evolutionary lineages, often associated with specific geographic regions along their natural areas of repartition in Europe, the Middle East, Western Asia and the African continent [4-8]. Three of these lineages are originally present in Europe: lineage A (the African lineage), spread through part of the Iberian Peninsula; C, widespread in southern and eastern Europe and M, found in the British Isles, north continental Europe, the Ural and some areas in Central Asia [9, 10].

The mitochondrial DNA (mtDNA) has been successfully used as a method to measure the genetic diversity and disentangle the evolutionary history and classification of *A. mellifera* [11, 12]. The mtDNA is commonly employed in phylogeography and genetic research because it exhibits no recombination, has a comparatively rapid evolutionary rate, a preserved genomic structure, and inheritance exclusively from one parent, which allows the clear tracing of lineages over generations [13, 14]. In particular, the intergenic non-coding region located between the cytochrome oxidase I (COX1) and cytochrome oxidase II (COX2) genes, known as 'COI-COII intergenic region', has been widely used in honeybee genetic studies (e.g., [4, 11, 15-17]). In addition to several point mutations, the COI-COII intergenic region has size variation due to the presence, absence or repetition of P and Q units. The P units has about 67 nt ( $P_0$ ) or 54 nt (P) due to a 13-nt internal deletion, while the Q element has about 200 nt. The presence and number of these P and Q units is used to distinguish African (A) and European (C and M) lineages: the C lineage lacks any P units and has one Q element; the A lineage has either  $P_0$  or P

units and one or two Q units and the M lineage has only P units and one to three Q elements [9, 10, 18].

The unique mtDNA sequences (known as haplotypes) obtained from honeybee samples has been used to identify their subspecies: A haplotypes occur in *A. m. iberiensis*, M haplotypes define *A. m. mellifera*, C1 classify *A. m. ligustica* and C2 *A. m. carnica* [1, 4, 5]. Previous works in Europe showed that A haplotypes are mostly restricted to Iberian peninsula, M haplotypes in central and norther Europe and C lineages in southern Europe, from Italy to Turkey [2]. These studied has also found evidence of human-mediated introgression at the lineage level in many European regions. The data is consistent with the fact that most beekeepers use preferentially the Italian *A. m. ligustica*, and the Carniolan *A. m. carnica* subspecies in their apiaries. For instance, studies in Belgium, Denmark, France, Germany, Poland, Sweden, and the UK, where only M lineages are native, revealed frequent introgression of C and O lineages [2]. The C branch (*A. m. ligustica*, *A. m. carnica*, *A. m. cecropia*) has spread from the Balkan and Italian Peninsulas to the northeastern coasts of the Mediterranean Sea and Central Europe. These introgressions result from the practice of long-distance translocation of subspecies for yield improvement and docility. Despite a centuries-long tradition of beekeeping and constant human interference, subspecies' specific genetic footprints are still preserved in some parts of Europe [4,33] but they are in a constant threat of hybridization, introgression and admixture which are detected in almost all analysed populations. Therefore, there is a growing interest to monitor the genetic integrity of the European *A. mellifera* subspecies that could be threatened by the human-mediated dispersion of non-native populations and lines.

The aim of this work was to perform a comprehensive genetic assessment of the genetic diversity present in Estonian honeybee populations. The analysis of mtDNA allowed the identification of the most widely circulating lineages among beekeepers. The new data can contribute to the implementation of conservation projects and to better understand the hybridization between subspecies.

## Methodology

### Sampling

A total of 156 honeybees were collected from 35 stationary apiaries in mainland Estonia and Muhu island during July 2023. Workers and drones were collected from 78 colonies, representing 154 samples. Two colonies were sampled only for workers, as drones were not found. Samples were collected from 13 counties in Estonia, the number of samples collected per county varied from 4 to 43 (Table 1). The number of samples collected per apiary varied from two to ten. The bees were stored in a solution of 70% ethanol until being processed in the laboratory. The DNA was extracted from the thoracic muscle mass obtained after opening the thorax with a sterilized scalpel. The DNA was extracted using the InviSorb® Spin Tissue Mini Kit (Invitex Molecular GmbH, Germany), according to the manufacturer's instructions. The extracted DNA was stored at  $-20\text{ }^{\circ}\text{C}$  for further analyses.

Table 1. Distribution of honeybee samples collected per county in Estonia.

County	N° of samples
Tartu county	43
Harju county	22
Pärnu county	17
Võru county	14
Lääne-Viru county	14
Saare county	10
Jõgeva county	10
Ida-Viru county	6
Viljandi county	4
Valga county	4
Rapla county	4
Lääne county	4
Järva county	4

### Amplification of the COI-COII intergenic region

The intergenic non-coding region of the mitochondrial DNA located between the cytochrome oxidase I (COX1) and cytochrome oxidase II (COX2) genes was selected for sequencing due to its wide use in honeybee genetic studies (Figure 1). The region is historically known as 'COI-COII intergenic region'. This section of the mtDNA was amplified by Polymerase Chain Reaction (PCR) using the following PCR primers previously described [4]:

BeeCOI3278F (COI\_Seq-F): 5'- ACCACCTCTAGATCATTACATTT -3'

BeeCOII4113R (COII\_Seq-R): 5'- AGGATGGAAGTGTTCATGAATGAA -3'

The PCR amplifications were performed on a SimpliAmp™ Thermal Cycler (Applied Biosystems, USA) in a 10 µL reaction mix containing 5 µL of Multiplex PCR Master Mix (Qiagen, Germany), 1 µL of primer mix (2 µM each), 2 µL of DNase, RNase and protease-free water and 2 µL of extracted DNA. The thermocycling conditions were: initial step at 95 °C for 15 min; 35 cycles at 94 °C for 30 s, 55 °C for 1 min 30 s, 72 °C for 1 min; with a final extension step at 72 °C for 10 min. The amplicons were separated by electrophoresis on a 2% agarose gel containing 1 X SYBR Safe DNA Gel Stain (Invitrogen, USA) at 120 V during 45 min and the fragments were visualized under UV light in a Dual LED Blue/White Light Transilluminator (Invitrogen).

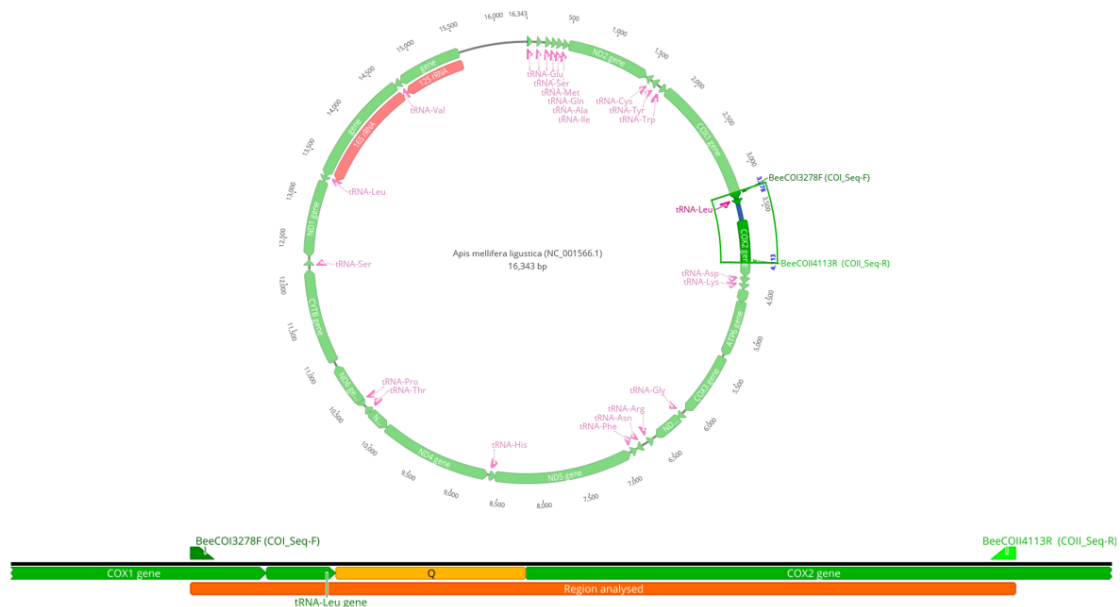


Figure 1. Schematic representation of the *Apis mellifera* mitochondrial DNA from the reference genome with accession number NC\_001566.1. The COI-COII intergenic region is highlighted by a green box. The PCR primers used to amplify the COI-COII intergenic region are indicated. The intergenic region of the reference genome includes a single Q element, typical of the mitochondrial lineage C.

## Sequencing of the COI-COII intergenic region

Amplified products (5 µL) were purified with ExoSAP-IT® Express (USB, Affymetrix, USA) according to the manufacturer's recommendations. Sequencing of purified samples (2.5 µL) was performed as follows: 10 µL reactions were prepared by combining 0.8 µL of Big Dye® Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems) with 0.8 µL of 10 µM primer, 1 µL of BigDye® Terminator v1.1 & v3.1 5X Sequencing Buffer and water DNase, RNase-free (Gibco, USA). Thermal cycler conditions were: 96°C for 2 min, 35 cycles at

96°C for 30 s, 50°C for 15 s and 60°C for 4 min and one final hold at 60°C for 10 min. Sequencing reaction products were purified using Sephadex™ G-50 Fine DNA Grade columns (GE Healthcare, United Kingdom) according to the manufacturer's recommendations. Purified samples were added to 12 µL Hi-Di™ formamide (Life Technologies, USA). Sequencing was performed in a Genetic Analyzer 3130xl sequencer (Applied Biosystems), according to the manufacturer's recommendations. Sequence analysis was performed using Sequencing Analysis software v6.0 (Applied Biosystems). The assembly of the resulting electropherograms was performed using Geneious Prime® 2022.1.1 (<https://www.geneious.com>). The final assemble was manually inspected to correct possible misalignments.

### **Identification of mitochondrial lineages**

The curated mtDNA sequences were aligned using the Muscle 3.8.425 [19] running in the Geneious Prime® software. The alignments were used to identify the sequences with extra copies of the two types of repeated, non-coding sequences, named P and Q. The variation in the number of P and Q elements allowed the classification of samples into the main C, M and A evolutionary lineages. The haplotype of each sample was identified using as reference the curated dataset with 934 full and verified COI-COII intergenic region sequences provided by Alburaki *et al.* [4]. A local database with the 934 sequences was built in the Geneious Prime® software and used for blast searches with the *blastn* algorithm under default score values. The final list of blast results was ordered by the % of identical sites and the haplotype of the query sample was obtained from the haplotype of the sequence with the highest value for the % of identical sites. Standard measures of genetic diversity were obtained using DNAsp 6.12.03 [20].

### **Phylogenetic analyses**

Phylogenies were built using the multiple sequence alignment with 156 COI-COII intergenic region sequences representing the mtDNA region from positions 3,339 to 4,063 of the reference *A. mellifera* mitochondrial genome (NC\_001566.1). The extra segment with ~250 bp only found in 14 individuals was removed from the alignment prior to the phylogenetic analyses. The median-joining network [21] was calculated using the Network V10.1.0.0 software (<http://www.fluxus-engineering.com>) using default parameters. The maximum likelihood (ML) phylogenetic tree was built with PhyML 3.0 [22], implemented in the ATGC bioinformatics platform (<http://www.atgc-montpellier.fr>). The HKY85 +I substitution model of protein evolution was selected with the Smart Model

Selection (SMS) v2.0 method implemented in PhyML [23], under the BIC (Bayesian Information Criterion). The branch support was evaluated with 100 bootstraps. The resulting phylogenetic tree was edited with FigTree v1.4.3 (<http://tree.bio.ed.ac.uk/software/figtree>).



## Results

### Main honeybee evolutionary lineages

The PCR amplification of the mtDNA COI-COII intergenic region using the PCR primers described in Figure 1 revealed two different product sizes (Figure 2). The sequencing of the PCR products showed that the longest amplicon had the PQQ or P<sub>0</sub>QQ structure whereas the shortest amplicon had the Q structure.

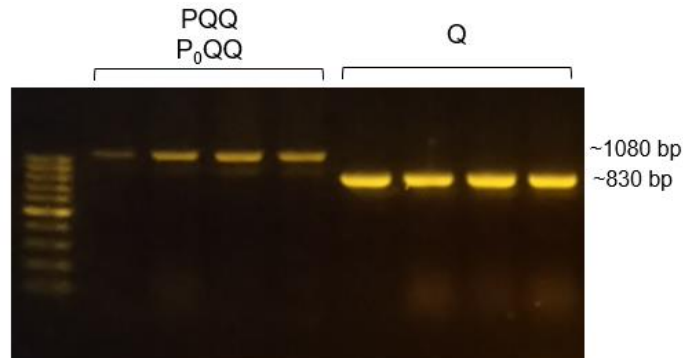


Figure 2. Electrophoretic separation of the COI-COII intergenic region amplified by PCR. The two main types of structure of the COI-COII intergenic region are shown (PQQ/P<sub>0</sub>QQ and Q).

The alignment of all honeybee mtDNA sequences revealed 14 cases with an insertion of around 250 nt in relation to the reference *A. mellifera* mitochondrial genome (Figure 3). Two samples (SCK040121 and SCK040122) have the structure P<sub>0</sub>QQ (1%) with a P element with no deletions and two copies of the Q element (Table 2). This structure is typical of the African mitochondrial lineage A. The remaining 12 samples (8%) have the structure PQQ, with a P element including a deletion of 13 nt. This structure is common in the western and northern European lineage M.

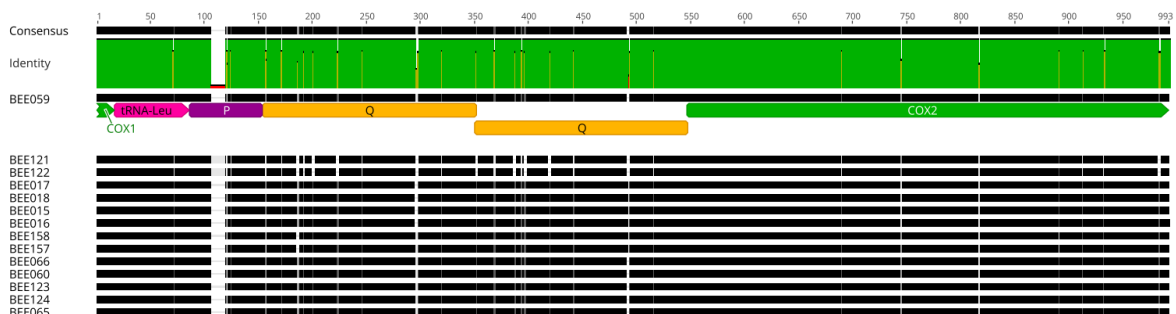


Figure 3. Multiple sequence alignment of the 14 samples with the PQQ and P<sub>0</sub>QQ structure in the COI-COII intergenic region.

The remaining 142 sequences (91%) have a single copy of the Q element (Table 2), typical of the southern and eastern European lineage C, as observed in the *Apis mellifera* reference genome (Figure 1).

Table 2. Classification of Estonian bee samples according to the COI-COII genetic structure and evolutionary lineage.

COI-COII genetic structure	Lineage	<i>n</i>	%
Q	C	142	91%
PQQ	M	12	8%
P <sub>0</sub> QQ	A	2	1%
	Total	156	

### Mitochondrial DNA haplotypes

The 156 samples were classified into 11 haplotypes using blast searches against the curated dataset of mtDNA sequences: C1a, C2c, C2d, C2e, C2j, C2s, C2w, M4, M4j, M4na and A2g (Table 3 and Supplementary Table S1).

Table 3. Classification of Estonian bee samples according to the mitochondrial DNA haplotype and subspecies.

COI-COII genetic structure	Lineage	Haplotype	Subspecies	<i>n</i>	%
Q	C	C1a	<i>Apis mellifera ligustica</i>	86	55.1%
		C2c	<i>Apis mellifera carnica</i>	10	6.4%
		C2d		27	17.3%
		C2e		8	5.1%
		C2j		2	1.3%
		C2s		8	5.1%
		C2w		1	0.6%
PQQ	M	M4	<i>Apis mellifera mellifera</i>	2	1.3%
		M4j		6	3.8%
		M4na		4	2.6%
P <sub>0</sub> QQ	A	A2g	<i>Apis mellifera iberiensis</i>	2	1.3%

The most common haplotype was C1a (55.1%), which characterizes the *A. m. ligustica* subspecies. This haplotype was found in 25 apiaries across Estonia (Supplementary Table S2). In addition to C1a, only haplotype C2d reached a relevant distribution (17.3%). The other haplotypes from the C lineage had a distribution of 6.4% (C2c) or lower. The haplotypes of the C2 type characterize the *A. m. carnica* subspecies. When

combined, C2 haplotypes comprise 35.9% of Estonian samples (Table 3). The haplotypes characterizing the *A. m. mellifera* subspecies (M4, M4j and M4na) represent 7.7% of the samples (Table 3) and were all collected in three apiaries (Harju County, Tartu maakond and Järva maakond). The single haplotype from the *A. m. iberiensis* subspecies (A2g) was found in a colony from the Jõgeva maakond, corresponding to only 1.3% of the samples.

Despite the smaller number of samples, *A. m. carnica* showed a higher genetic diversity than *A. m. ligustica* (Table 4). The presence of six haplotypes in *A. m. carnica* and only two in *A. m. ligustica* explains the higher diversity values in *A. m. carnica*. The genetic diversity of the three *A. m. mellifera* haplotypes is comparable to that of *A. m. carnica* (Table 4).

Table 4. Diversity measures for the *Apis mellifera* subspecies observed in Estonia based on the COI-COII intergenic region.

Lineage	Subspecies	<i>n</i>	N° of polymorphic sites	N° of haplotypes	Haplotype diversity	Nucleotide diversity	Average n° of nucleotide differences
C	<i>A. m. ligustica</i>	86	1	2	0.243	0.00034	0.243
	<i>A. m. carnica</i>	56	8	6	0.608	0.00181	1.308
M	<i>A. m. mellifera</i>	12	3	3	0.667	0.00184	1.333
A	<i>A. m. iberiensis</i>	2	0	1	0	0	0
	Complete dataset	156	19	11	0.646	0.00386	2.781

## Phylogenetic analyses

The phylogenetic network of mtDNA haplotypes revealed three clearly distinct clusters of haplotypes separated by eight mutations: *A. m. mellifera*, *A. m. iberiensis* and *A. m. ligustica* + *A. m. carnica* (Figure 4). The difference among the three clusters is compatible with the existence of difference subspecies in *A. mellifera*. However, the separation of *A. m. ligustica* and *A. m. carnica* is not evident in the Estonian samples. Haplotypes C2c and C1a are just one mutation apart. The C2 haplotypes form a starlike pattern expanding from C2d, which is possible one of the ancestral haplotypes in this cluster. In fact, samples classified as C2d were found to have two haplotypes separated by one mutation at the centre of the network, in line with its ancestral state. Similarly, C1a samples revealed two haplotypes in the median-joining network (Figure 4). In both cases,

it is possible that some of the samples could be further classified into new haplotypes. It is also by this reason that the network shows 13 haplotypes instead of the 11 haplotypes identified by blast analyses (Table 3). The three *A. m. mellifera* haplotypes (M4, M4j and M4na) are separated by at least three mutations suggesting they diverge already some time ago.

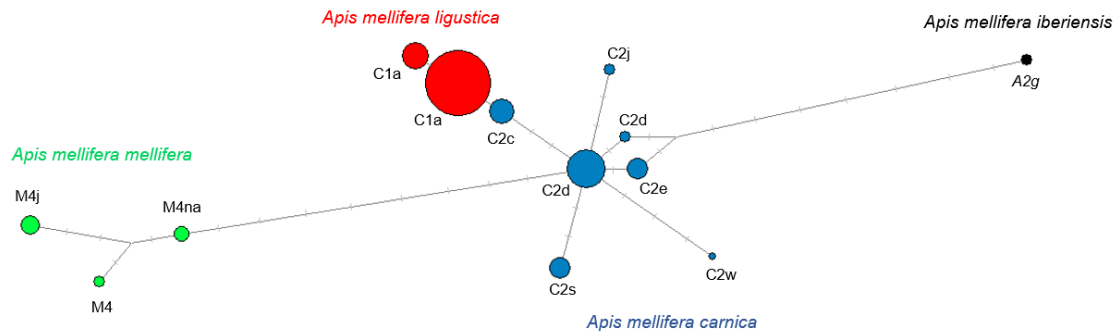


Figure 4. Median-joining network built with the alignment of 156 COI-COII intergenic region sequences of Estonian honeybees. The area of the circles is proportional to the frequency of sequences. Haplotypes are identified and coloured according to the *A. mellifera* subspecies.

The maximum likelihood (ML) phylogenetic tree supported the median-joining network analysis by separating all samples according to the haplotype classification (Figure 5). The *A. m. ligustica* and *A. m. carnica* samples cluster together, but with no overlap of haplotypes between both groups. The *A. m. mellifera* and *A. m. iberiensis* haplotypes form a separated cluster, although *A. m. iberiensis* branches out from *A. m. carnica*.

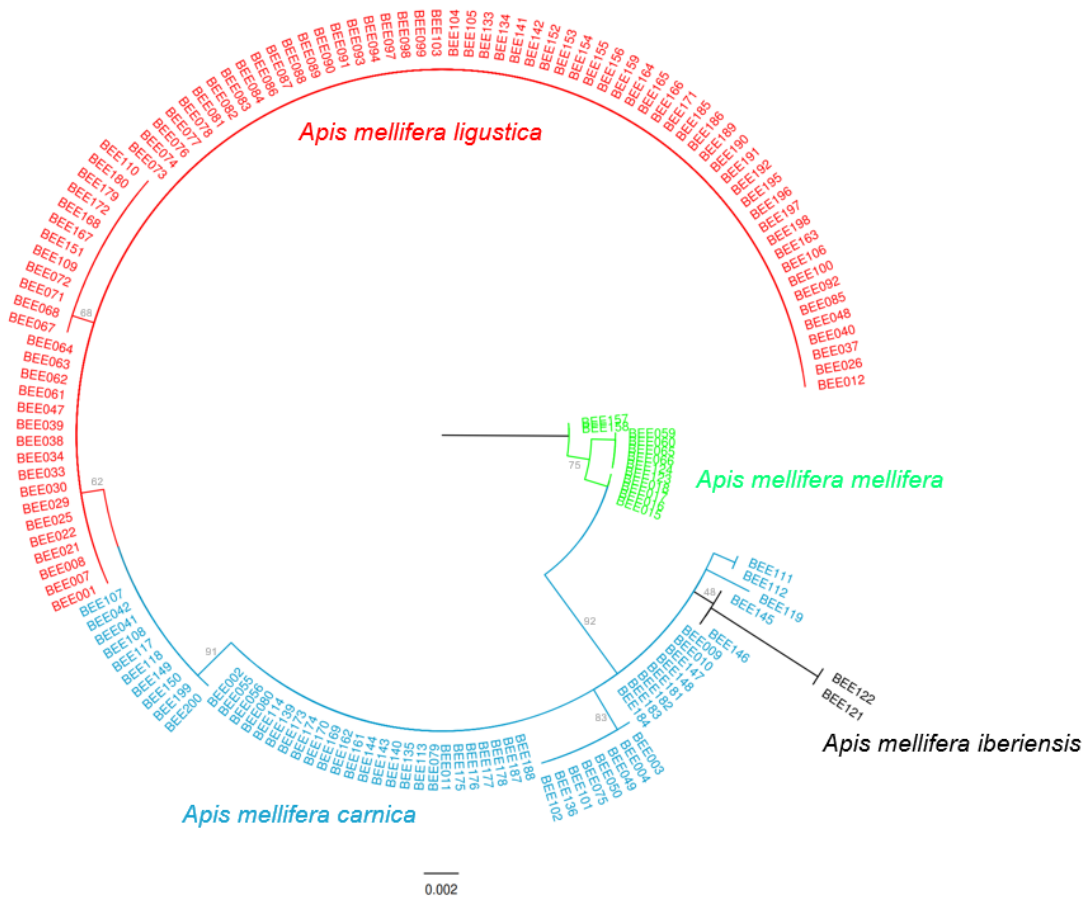


Figure 5. Maximum likelihood (ML) phylogenetic tree built with the alignment of 156 COI-COII intergenic region sequences of Estonian honeybees. The branch support was estimated with 100 bootstraps. The scale bar indicates substitutions per site.

### Beekeepers' classifications

The genetic analyses revealed six cases of workers and drones from the same colony with different mtDNA sequences (Table 5). Although one should expect equal mtDNA sequences due to their descent from the same female, it is possible the occurrence of *de novo* mutations through the multiple mtDNA replications in the development of an organism. The cases of workers and drones diverging by just a few mutations (one or even two) could be explained by such process. However, the cases with three and five mutations are unlikely to have occurred *de novo* in an individual. It is possible that drones from different colonies were sampled or that some misclassification of samples has occurred.

Table 5. List of workers and drone samples from the same colony showing different mtDNA sequences for the COI-COII intergenic region with ~755 nt.

Samples from the same colony with different mtDNA sequence	Number of nt differences	% identical sites
SCK040001 vs SCK040002	3	99.6
SCK040011 vs SCK040012	3	99.6
SCK040075 vs SCK040076	5	99.3
SCK040135 vs SCK040136	2	99.7
SCK040151 vs SCK040152	1	99.9
SCK040171 vs SCK040172	1	99.9

The genetic classification of subspecies using the COI-COII intergenic region not always matched the subspecies reported by the beekeepers (Table 6). It should be noted that the mtDNA lineage may not always reflect the morphology of the bee, particularly if recent crosses between individuals of different lineages may have occurred. The most common discordance between the mtDNA and the beekeeper's classifications regards *A. m. ligustica* and *A. m. carnica*. Half the samples reported as *A. m. carnica* by beekeepers were classified as *A. m. ligustica* by the mtDNA analysis, and the opposite scenario occurred in 28% of the cases. The genetic proximity between these two subspecies, as noticed in the phylogenetic analyses (Figures 4 and 5), may explain the difficulty in separating both subspecies. In any case, 66% of *A. m. ligustica* samples identified by beekeepers were supported by the mtDNA data (Table 6). The two samples of *A. m. iberiensis* detected using mtDNA have been reported as *A. m. ligustica* by the beekeeper. There was a concordance for 60% of the cases between the beekeeper and mtDNA classification for *A. m. mellifera*. The 12 cases of bees reported as Buckfast have a mtDNA haplotype typical of *A. m. carnica*.

Table 6. Comparison of the honeybee subspecies classification according to beekeepers and the mitochondrial DNA haplotypes.

Subspecies reported by beekeeper	<i>n</i>	Subspecies according to mtDNA haplotypes	<i>n</i>	%
<i>Apis mellifera ligustica</i>	97	<b><i>A. m. ligustica</i></b>	<b>64</b>	<b>66%</b>
		<i>A. m. carnica</i>	27	28%
		<i>A. m. mellifera</i>	4	4%
		<i>A. m. iberiensis</i>	2	2%
<i>Apis mellifera carnica</i>	40	<i>A. m. ligustica</i>	20	50%
		<b><i>A. m. carnica</i></b>	<b>16</b>	<b>40%</b>
		<i>A. m. mellifera</i>	4	10%
<i>Apis mellifera mellifera</i>	10	<i>A. m. ligustica</i>	2	20%
		<i>A. m. carnica</i>	2	20%
		<b><i>A. m. mellifera</i></b>	<b>6</b>	<b>60%</b>
<i>Buckfast</i>	12	<i>A. m. carnica</i>	12	100%

## Conclusions

1. The honeybee population of Estonia includes individuals belonging to the three main mitochondrial lineages [17]:
  - C (91%); subspecies native to southern and eastern Europe
  - M (8%); subspecies native to western and northern Europe
  - A (1%); subspecies native to Africa and Iberian Peninsula.
2. The Estonian honeybee population is mostly (91%) comprised of two subspecies of the lineage C [1]:
  - Apis mellifera ligustica* (55.1%): Italian honeybee; subspecies native to Italy
  - Apis mellifera carnica* (35.9%): Carniolan honeybee; subspecies native to Slovenia, Bulgaria, Poland, Austria, Croatia, Bosnia and Herzegovina, Serbia, Hungary and Romania.
3. Only 7.7% of Estonian bees analysed here belong to the native *Apis mellifera mellifera* subspecies (European dark honeybee).
4. The haplotype diversity of the honeybee population in Estonia is comparable to that found in other world regions. For example, the number of different mtDNA haplotypes observed in Estonian bees (11 haplotypes in 156 sequences or 79 colonies) is within the range of what was observed in recent studies from other regions using the same mtDNA segment: Serbia (9 haplotypes in 241 samples) [24], USA (27 haplotypes in 1,063 samples) [4], Peru (24 haplotypes in 512 samples) [16], northern Poland, Hungary and Romania (45 haplotypes in 444 samples) [15] and 12 African countries (84 haplotypes in 1184 samples) [8].
5. Samples from *A. m. ligustica* belong to only two different haplotypes suggesting a low genetic diversity for individuals of this subspecies. This result can be explained either by the recurrent introduction of individuals from the same lineage or the local dispersion from a single early introduction in Estonia.
6. Samples from *A. m. carnica* showed a higher genetic diversity than *A. m. ligustica*, with seven different mtDNA haplotypes. This result suggest that multiple individuals of this species may have been introduced in Estonia.

7. Samples from *A. m. mellifera* are grouped into three different haplotypes found in three separated apiaries located in Harju County, Tartu maakond and Järva maakond. Considering that this subspecies is native to Estonia and that the apiaries are in different regions, it is possible that the individuals represent local native lineages that were collected by the beekeepers. Nevertheless, the independent introduction of *A. m. mellifera* from other regions of its natural distribution cannot be excluded with the present dataset.
8. The six cases of different mtDNA sequences between workers and drones of the same colony require further investigation. The cases with a few mutations of difference could be the result of new mutations within each individual, but the cases with several mutations could represent some sample processing error. The analysis of additional samples collected at the same time will clarify the situation.
9. There was a discordance in 46 % of samples between the classification of honeybees' subspecies by beekeepers and by the mtDNA analyses. The discordance can result from:
  - a) The existence of recent hybridization. Mitochondria are inherited exclusively from the female, meaning that only one parent's mitochondrial lineage is represented in the offspring. In hybridization events between two different subspecies, if the female belongs to one species and the male to another, the mtDNA of the hybrid will only reflect the maternal lineage. As a result, it may not accurately represent the genetic contribution of the other parent or the true hybrid nature of the individual.
  - b) The low genetic difference between *A. m. ligustica* and *A. m. carnica*. Since 30% of the samples with discordance in the classifications occurs between these species, it is possible that the similarity between the two species may lead to misclassifications using morphology alone.
10. All Buckfast honeybees had *A. m. carnica* maternal lineage, which is unexpected considering the origin of this breed using queens from Italy *A. m. ligustica* [25]. This result may suggest additional crossings in this Buckfast lineage.
11. Overall, the Estonian honeybee population is mainly comprised of commercial C-lineage honeybees. The native honeybee subspecies (*A. m. mellifera*) is rare,



despite representing a potential reservoir of unique combinations of genes adapted to local conditions. The level of genetic diversity is similar to other world regions, suggesting similar breeding practices.

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## Supplementary Material

Supplementary Table S1. Classification of the 156 samples of Estonian honeybees according to the COI-COII intergenic region of the mitochondrial DNA.

Sample ID	Honeybee type/gender	Subspecies	COI-COII genetic structure	Lineage	Haplotype
SCK040065	worker	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040066	drone	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040059	worker	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040060	drone	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040123	worker	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040124	drone	<i>A. m. mellifera</i>	PQQ	M	M4j
SCK040181	worker	<i>A. m. carnica</i>	Q	C	C2e
SCK040182	drone	<i>A. m. carnica</i>	Q	C	C2e
SCK040109	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040110	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040107	worker	<i>A. m. carnica</i>	Q	C	C2c
SCK040108	drone	<i>A. m. carnica</i>	Q	C	C2c
SCK040167	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040168	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040071	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040072	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040105	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040106	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040171	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040172	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040177	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040178	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040135	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040136	drone	<i>A. m. carnica</i>	Q	C	C2s
SCK040175	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040176	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040173	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040174	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040139	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040140	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040169	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040170	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040033	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040034	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040029	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040030	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040143	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040144	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040141	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040142	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040159	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040165	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040166	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040155	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040156	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040157	worker	<i>A. m. mellifera</i>	PQQ	M	M4
SCK040158	drone	<i>A. m. mellifera</i>	PQQ	M	M4

SCK040153	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040154	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040025	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040026	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040037	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040038	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040187	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040188	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040185	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040186	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040179	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040180	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040077	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040078	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040089	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040090	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040085	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040086	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040081	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040082	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040083	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040084	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040091	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040092	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040087	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040088	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040073	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040074	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040161	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040162	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040121	worker	<i>A. m. iberiensis</i>	P0QQ	A	A2g
SCK040122	drone	<i>A. m. iberiensis</i>	P0QQ	A	A2g
SCK040163	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040164	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040075	worker	<i>A. m. carnica</i>	Q	C	C2s
SCK040076	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040021	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040022	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040039	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040040	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040017	worker	<i>A. m. mellifera</i>	PQQ	M	M4na
SCK040018	drone	<i>A. m. mellifera</i>	PQQ	M	M4na
SCK040015	worker	<i>A. m. mellifera</i>	PQQ	M	M4na
SCK040016	drone	<i>A. m. mellifera</i>	PQQ	M	M4na
SCK040061	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040064	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040063	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040062	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040119	worker	<i>A. m. carnica</i>	Q	C	C2w
SCK040117	worker	<i>A. m. carnica</i>	Q	C	C2c
SCK040118	drone	<i>A. m. carnica</i>	Q	C	C2c
SCK040067	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040068	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040151	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040152	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040103	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040104	drone	<i>A. m. ligustica</i>	Q	C	C1a

SCK040097	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040098	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040093	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040094	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040099	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040100	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040195	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040196	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040197	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040198	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040191	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040192	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040183	worker	<i>A. m. carnica</i>	Q	C	C2e
SCK040184	drone	<i>A. m. carnica</i>	Q	C	C2e
SCK040041	worker	<i>A. m. carnica</i>	Q	C	C2c
SCK040042	drone	<i>A. m. carnica</i>	Q	C	C2c
SCK040009	worker	<i>A. m. carnica</i>	Q	C	C2e
SCK040010	drone	<i>A. m. carnica</i>	Q	C	C2e
SCK040003	worker	<i>A. m. carnica</i>	Q	C	C2s
SCK040004	drone	<i>A. m. carnica</i>	Q	C	C2s
SCK040199	worker	<i>A. m. carnica</i>	Q	C	C2c
SCK040200	drone	<i>A. m. carnica</i>	Q	C	C2c
SCK040047	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040048	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040145	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040146	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040079	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040080	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040101	worker	<i>A. m. carnica</i>	Q	C	C2s
SCK040102	drone	<i>A. m. carnica</i>	Q	C	C2s
SCK040113	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040114	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040111	worker	<i>A. m. carnica</i>	Q	C	C2j
SCK040112	drone	<i>A. m. carnica</i>	Q	C	C2j
SCK040011	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040012	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040001	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040002	drone	<i>A. m. carnica</i>	Q	C	C2d
SCK040007	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040008	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040133	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040134	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040149	worker	<i>A. m. carnica</i>	Q	C	C2c
SCK040150	drone	<i>A. m. carnica</i>	Q	C	C2c
SCK040147	worker	<i>A. m. carnica</i>	Q	C	C2e
SCK040148	drone	<i>A. m. carnica</i>	Q	C	C2e
SCK040189	worker	<i>A. m. ligustica</i>	Q	C	C1a
SCK040190	drone	<i>A. m. ligustica</i>	Q	C	C1a
SCK040049	worker	<i>A. m. carnica</i>	Q	C	C2s
SCK040050	drone	<i>A. m. carnica</i>	Q	C	C2s
SCK040055	worker	<i>A. m. carnica</i>	Q	C	C2d
SCK040056	drone	<i>A. m. carnica</i>	Q	C	C2d

Table S1. Distribution of the 156 samples of Estonian honeybees according to beekeeper.

Beekeeper	Sample ID	Honeybee type/gender	Apiary	Haplotype	Subspecies (mtDNA)	Expected race/subspecies of honeybees
BK_01	SCK040109	worker	AP_01	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040110	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040107	worker		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040108	drone		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_02	SCK040195	worker	AP_02	C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040196	drone		C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040197	worker		C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040198	drone		C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040191	worker		C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040192	drone		C1a	<i>Apis mellifera ligustica</i>	unknown ( <i>Apis mellifera ligustica</i> (italian honey bee)
BK_03	SCK040017	worker	AP_03	M4na	<i>Apis mellifera mellifera</i>	<i>Apis mellifera mellifera</i> (european dark bee)
	SCK040018	drone		M4na	<i>Apis mellifera mellifera</i>	<i>Apis mellifera mellifera</i> (european dark bee)
	SCK040015	worker		M4na	<i>Apis mellifera mellifera</i>	<i>Apis mellifera mellifera</i> (european dark bee)
	SCK040016	drone		M4na	<i>Apis mellifera mellifera</i>	<i>Apis mellifera mellifera</i> (european dark bee)
BK_04	SCK040145	worker	AP_04	C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040146	drone		C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040079	worker		C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040080	drone		C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040101	worker		C2s	<i>Apis mellifera carnica</i>	Buckfast
	SCK040102	drone		C2s	<i>Apis mellifera carnica</i>	Buckfast
	SCK040113	worker		C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040114	drone		C2d	<i>Apis mellifera carnica</i>	Buckfast
	SCK040111	worker		C2j	<i>Apis mellifera carnica</i>	Buckfast
	SCK040112	drone		C2j	<i>Apis mellifera carnica</i>	Buckfast
BK_05	SCK040047	worker	AP_05	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040048	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_06	SCK040065	worker	AP_06	M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040066	drone		M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040059	worker		M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040060	drone		M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040123	worker		M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040124	drone		M4j	<i>Apis mellifera mellifera</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_07	SCK040033	worker	AP_07	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040034	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040029	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)

	SCK040030	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_08	SCK040167	worker	AP_08	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040168	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040071	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040072	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040149	worker	AP_09	C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carpathica</i>
	SCK040150	drone		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carpathica</i>
	SCK040147	worker		C2e	<i>Apis mellifera carnica</i>	Unknown
	SCK040148	drone		C2e	<i>Apis mellifera carnica</i>	Unknown
	SCK040189	worker	AP_08	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera mellifera</i> (european dark bee)?
	SCK040190	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera mellifera</i> (european dark bee)?
BK_09	SCK040077	worker	AP_10	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040078	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040089	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040090	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040085	worker	AP_11	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040086	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040081	worker	AP_12	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040082	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040083	worker	AP_11	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040084	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040091	worker	AP_10	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040092	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040087	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040088	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040073	worker	AP_11	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040074	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_10	SCK040143	worker	AP_13	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> / <i>Apis mellifera mellifera</i>
	SCK040144	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> / <i>Apis mellifera mellifera</i>
	SCK040141	worker	AP_14	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040142	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040159	worker	AP_13	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040165	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040166	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040155	worker	AP_15	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040156	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040157	worker	AP_13	M4	<i>Apis mellifera mellifera</i>	<i>Apis mellifera ligustica</i> , <i>Apis mellifera mellifera</i>
	SCK040158	drone		M4	<i>Apis mellifera mellifera</i>	<i>Apis mellifera ligustica</i> , <i>Apis mellifera mellifera</i>
	SCK040153	worker	AP_16	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)



	SCK040154	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
BK_11	SCK040105	worker	AP_17	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040106	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040171	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040172	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
BK_12	SCK040011	worker	AP_18	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040012	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040001	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040002	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040007	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
BK_13	SCK040061	worker	AP_19	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040064	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040063	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040062	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
BK_14	SCK040133	worker	AP_20	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040134	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
BK_15	SCK040183	worker	AP_21	C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040184	drone		C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040041	worker		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040042	drone		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040009	worker		C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
BK_16	SCK040010	drone		C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica (carniolan honey bee)</i>
	SCK040103	worker	AP_22	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040104	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040097	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040098	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040093	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040094	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040099	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
SCK040100	drone	C1a		<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>	
BK_17	SCK040177	worker	AP_23	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040178	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040135	worker	AP_24	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040136	drone		C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040175	worker	AP_23	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040176	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040173	worker		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040174	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>

	SCK040139	worker	AP_24	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040140	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040169	worker		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040170	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_18	SCK040119	worker	AP_25	C2w	<i>Apis mellifera carnica</i>	unknown
	SCK040117	worker		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040118	drone		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_19	SCK040021	worker	AP_26	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040022	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040039	worker	AP_27	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040040	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_20	SCK040049	worker	AP_28	C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040050	drone		C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040055	worker		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040056	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
BK_21	SCK040003	worker	AP_29	C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> or <i>Apis mellifera carnica</i>
	SCK040004	drone		C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> or <i>Apis mellifera carnica</i>
	SCK040199	worker		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> or <i>Apis mellifera carnica</i>
	SCK040200	drone		C2c	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> or <i>Apis mellifera carnica</i>
BK_22	SCK040067	worker	AP_30	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040068	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040151	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040152	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_23	SCK040181	worker	AP_31	C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040182	drone		C2e	<i>Apis mellifera carnica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_24	SCK040187	worker	AP_32	C2d	<i>Apis mellifera carnica</i>	Buckfast (Tenuta Ritiro F1)
	SCK040188	drone		C2d	<i>Apis mellifera carnica</i>	Buckfast (Tenuta Ritiro F1)
	SCK040185	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> , <i>Apis mellifera carpathica</i>
	SCK040186	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> , <i>Apis mellifera carpathica</i>
	SCK040179	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
	SCK040180	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera carnica</i> (carniolan honey bee)
BK_25	SCK040161	worker	AP_33	C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040162	drone		C2d	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040121	worker	AP_34	A2g	<i>Apis mellifera iberiensis</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040122	drone		A2g	<i>Apis mellifera iberiensis</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040163	worker	AP_33	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040164	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040075	worker		C2s	<i>Apis mellifera carnica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)
	SCK040076	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica</i> (italian honey bee)

BK_26	SCK040025	worker	AP_35	C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040026	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040037	worker		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>
	SCK040038	drone		C1a	<i>Apis mellifera ligustica</i>	<i>Apis mellifera ligustica (italian honey bee)</i>