Characterisation and tracking changes of morphological characteristics in honey bee, *Apis mellifera*, colonies

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Abstract

Measuring morphological characteristics of honey bee, *Apis mellifera*, at different times is important to assess the degree of race purity, to predict colony productivity, and to understand environmental impacts on honey bees. The study aims to characterise current honey bees of Northwest Egypt, namely El-Behera governorate, and to track occurred changes to their morphological characteristics since 2007. Samples of honey bee workers were collected from four districts in El-Behera governorate. Nine body and six wing characteristics were measured. Morphological characteristics of honey bees belonging to these districts were previously measured during 2007. To track changes, a comparison between new (2014) and previous (2007) measurements of morphological characteristics was done. The current bees of El-Behera are slightly larger than those of 2007, emphasising no reduction in colonies productivity. There is evidence that environmental factors have not caused much stress on the colonies since 2007. The results of this study are useful in providing understanding of the current state of bees in Egypt, and the likely improvement of future beekeeping.

Introduction

Morphological characteristics of honey bees, *Apis mellifera*, are very important to follow changes of honey bee colonies over time (Abou-Shaara *et al.*, 2012b), to investigate hybridisation with other subspecies (Radloff *et al.*, 2003), to characterise honey bee subspecies (Meixner *et al.*, 2007), to study symmetry of body parts and to discriminate between subspecies (Abou-Shaara & Al-Ghamdi, 2012). Morphological studies on honey bees can be divided into two basic categories: standard (using wing and body characteristics) and geometric morphometrics (using coordinates of wing points). Recently these two categories have been intensively reviewed by Abou-Shaara *et al.* (2013) and Abou-Shaara (2013).

It is well known that different morphological characteristics of honey bees (*e.g.*, wings and legs) are correlated with honey production (Milne & Pries, 1984; Waddington, 1989; Kolmes & Sam, 1991; Edriss *et al.*, 2002; Mostajeran *et al.*, 2006). Basically, bees with large body characteristics are better than those with smaller ones in regard to honey production, but in regard to tolerance to thermal stress the opposite has been found (Abou-Shaara *et al.*, 2012a). Hence, measuring morphological characteristics over time is important to detect the occurrence of any changes to honey bee colonies in regard to subspecies purity, colonies productivity, and tolerance to environmental factors.

In the El-Behera governorate, Egypt, Abou-Shaara *et al.* (2012b) have studied morphological characteristics of managed honey bee colonies belonging to different districts during two successive years 2006 and 2007. They found a decline in most measured characteristics in 2007 compared to 2006. It has been hypothesised that the decline in morphological characteristics would be continued over time. The objectives of this study are therefore, to test this hypothesis, by the characterisation of current honey bees of the El-Behera governorate, and tracking the actual changes to morphological characteristics.

Materials and methods

Sampling

Samples of honey bee workers were collected during autumn 2014 from 4 districts (Damanhour, El-Mahmoudia, El-Dalangat and Hosh Esa) as shown in Figure 1. Total of 32 colonies were sampled (8 colonies/apiary per district), and 20 workers were collected from each colony (total of 640 workers). The collected samples were kept in ethyl alcohol 70% till analysis.

Taking measurements

Collected workers were dissected using forceps to separate heads, wings and legs. The separated body parts were then scanned using

Canon scanner (k10352, CanoScan LiDE 110, Vietnam) at 1200 dpi. The images obtained were subsequently analysed in Adobe Photoshop® (Adobe Systems Inc., San Jose, CA, USA) according to Abou-Shaara *et al.* (2011) to measure: head width (HW), fore wing length (FWL) and width (FWW), hind wing length (HWL) and width (HWW), femur length (FL), tibia length (TL), and basetarsus length (BL) and width (BW), and cubital index (CI) which was calculated as distance A/distance B. Some wing angles (A4, D7, J16, K19 and Q21) were measured using imageJ 1.46 program.

Characterisation

Firstly, a principal component analysis was carried out to compare the four districts using R package 3.2.1. Then means of measured characteristics for studied districts were compared by Duncan s multiple range (α=0.05) using SAS 9.1.3 (2004). The measured characteristics for the studied districts were then compared with means presented in Abou-Shaara (2009) for native honey bees of Egypt (*Apis mellifera lamarckii*), Carniolan honey bees (*A.m. carnica*) and Italian honey bees (*A.m.ligustica*).

Track changes of measured characteristics

To track changes of honey bee characteristics at El-Behera governorate, morphological characteristics for the studied districts were compared with the most recent data of morphological characteristics during 2007 (Abou-Shaara *et al.*, 2012b) for the same districts. A principal component analysis starting from means of 2007 and 2014 was performed to compare years using R package 3.2.1.

Results

Characterisation

Significant differences were found among studied districts in the measured characteristics. Only HW showed significant differences among all the studied districts while the other characters showed significant differences only between 2 or 3 districts. The highest means for 6 characters were found in colonies belonging to Damanhour district while the other districts had the highest means
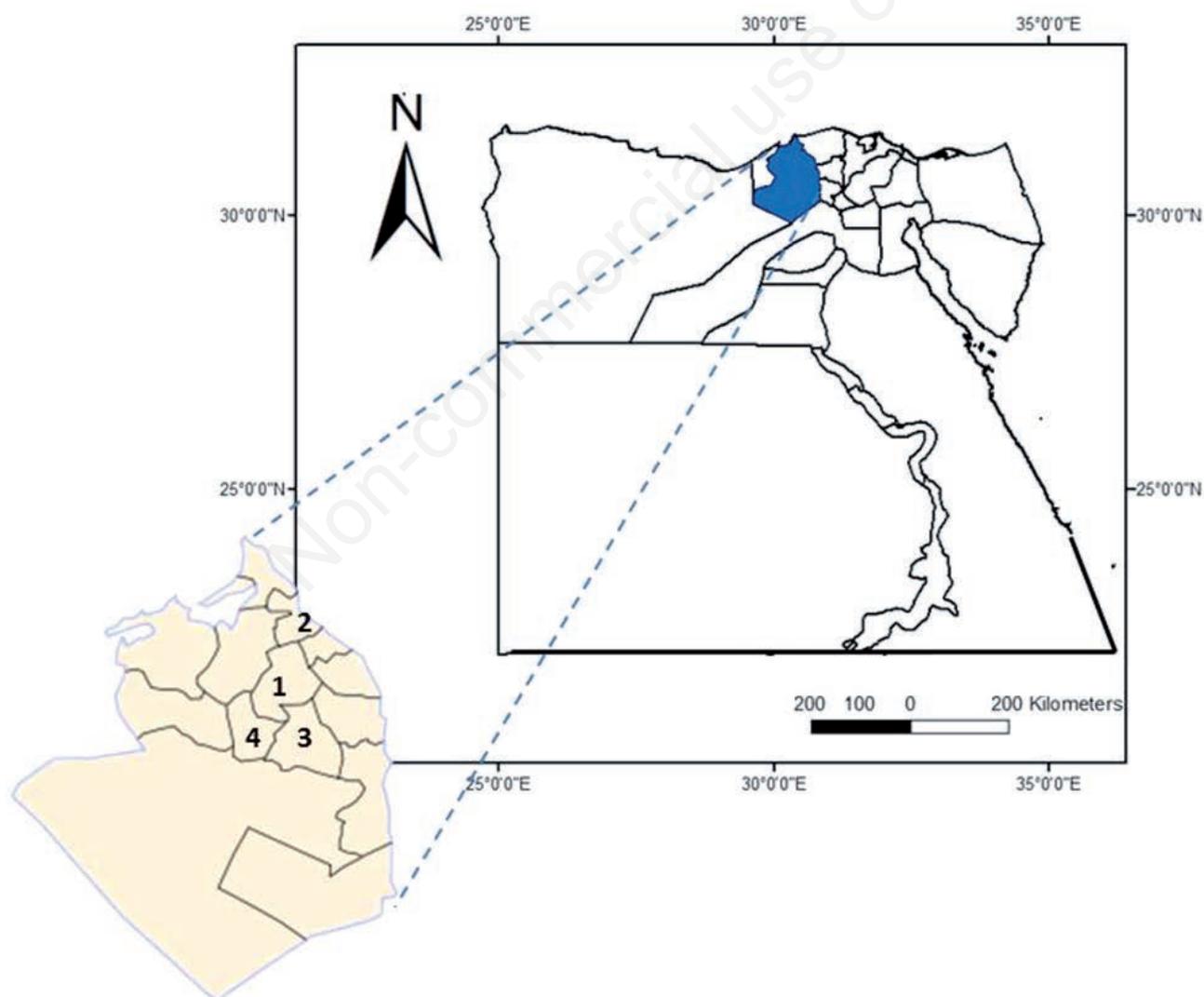
Figure 1. Location of El-Behera governorate in Egypt (located between 30° 36' 36" N and 30° 25' 48" E with total area of 9826 km²), and studied districts (1, Damanhour; 2, El-Mahmoudia; 3, El-Dalangat; 4, Hosh Esa).

only for a lower number of characters per district (Table 1). The overall means for measured characteristics are higher than those of native honey bees of Egypt (*A.m. lamarckii*) by 0.45, 0.26, 0.05 and 0.62 mm for FWL, FWW, BL and CI, respectively. Carniolan honey bees (*A.m. carnica*) have higher means than El-Behera bees by 0.49, 0.15, 0.57, 0.18, 0.41 and 0.06 mm for FWL, FWW, HWL, HWW, BL and BW, respectively. Italian honey bees (*A.m.ligustica*), also, have higher means than El-Behera bees by 0.51, 0.18 and 0.07 mm for FWL, FWW and BL, respectively. El-Behera honey bees have higher means than Carniolan and Italian honey bees only in CI value, measuring 0.43 and 0.32, respectively.

When HW was plotted against TL using SYSTAT 13 (Figure 2), a separation for studied colonies was obtained with few overlapping. However, it was not possible to separate the studied colonies into well distrinct clusters when all characters were included in a principal component analysis, even if some separation of cluster is apparent (Figure 3). The overlap between colonies belonging to the studied districts suggests a high degree of hybridisation.

Track changes of measured characteristics

As presented in Table 2, FWW, FL and TL showed increases in all districts while BL showed decrease in all districts. CI showed decrease in Damanhour only. The other characteristics showed increase only in Damanhour except HWW which increased only in El-Dalangat. The increase ranged from 0.03 to 0.54 while the decrease ranged from 0.01 to 0.4. The increasing in characteristics relate to body size reflect that honey bees of El-Behera are currently larger than the past (Figure 4).

As shown in Figure 5, five characteristics showed increases in their overall means during 2014 compared to 2007 (namely; FWW, FL, TL, and BW, and CI) with difference of 0.11, 0.07, 0.1 and 0.01mm, and 0.2, respectively. Three characteristics (FWL, HWL and BL) showed decreases in 2014 compared to 2007 of 0.04, 0.02 and 0.09mm while HWW showed no change between 2007 and 2014. Principal component analysis emphasised a clear separation between means of 2007 and

2014 (Figure 6), confirming some modification of morphometric characteristics of populations in time.

Discussion

Characterisation

The measured means of El-Behera bees from different districts showed that current bees are larger than native Egyptian bees but
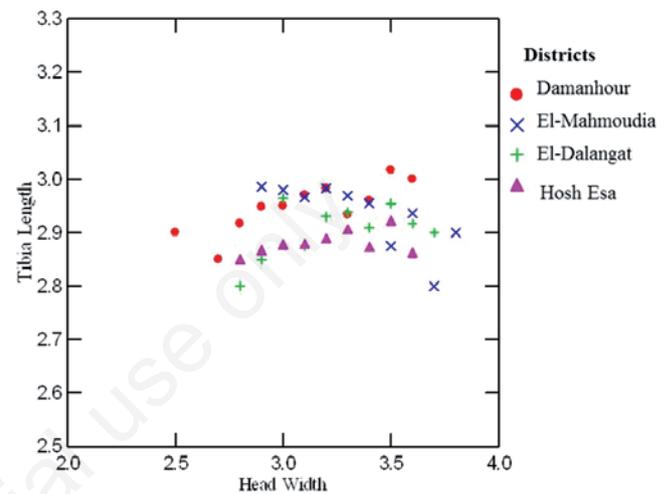
Figure 2. Means of tibia length against means of head width, grouped by studied districts.

Table 1. Means±standard deviation of measured characteristics for studied districts. All characteristics are in mm except angles in degrees, and cubital index without units.

Characteristics	Districts				
	Damanhour	El-Mahmoudia	El-Dalangat	Hosh Esa	Overall mean
Head width	3.09±0.21[d]	3.22±0.21[c]	3.32±0.19[a]	3.27±0.18[b]	3.22±0.09
Fore wing length	8.74±0.20[a]	8.70±0.17[ab]	8.67±0.19[bc]	8.64±0.19[c]	8.68±0.04
Fore wing width	3.06±0.09[a]	3.06±0.10[a]	3.02±0.10[b]	3.02±0.11[b]	3.04±0.02
Hind wing length	6.09±0.20[b]	6.08±0.17[b]	6.14±0.15[a]	6.01±0.21[c]	6.08±0.05
Hind wing width	1.70±0.13[b]	1.75±0.18[a]	1.78±0.13[a]	1.77±0.18[a]	1.75±0.03
Femur length	2.34±0.13[a]	2.32±0.14[a]	2.25±0.17[b]	2.25±0.12[b]	2.29±0.04
Tibia length	2.95±0.11[a]	2.96±0.10[a]	2.92±0.11[b]	2.88±0.09[c]	2.92±0.03
Basetarsus length	2.01±0.11[b]	2.02±0.10[b]	1.98±0.09[c]	2.04±0.09[a]	2.01±0.02
Basetarsus width	1.15±0.10[a]	1.12±0.07[b]	1.09±0.10[c]	1.10±0.06[c]	1.11±0.02
Ditsance A	0.51±0.05[b]	0.52±0.06[b]	0.51±0.06[b]	0.55±0.06[a]	0.52±0.02
Distance B	0.19±0.03[a]	0.19±0.05[a]	0.19±0.04[a]	0.19±0.04[a]	0.19±0.00
Cubital index	2.79±0.97[a]	2.96±1.19[ab]	2.95±1.17[ab]	3.12±1.17[a]	2.95±0.13
Angle A4	34.56±3.27[a]	35.20±3.30[a]	35.14±3.26[a]	33.25±2.51[b]	34.53±0.90
Angle D7	93.72±4.18[b]	92.50±4.53[c]	93.24±4.49[bc]	94.77±3.82[a]	93.55±0.95
Angle J16	94.84±6.73[b]	96.99±6.76[a]	95.09±6.39[b]	95.25±6.23[b]	95.54±0.97
Angle K19	79.01±4.50[a]	77.98±4.07[b]	77.70±4.05[b]	77.18±4.15[b]	77.96±0.76
Angle Q21	35.12±2.66[a]	34.16±2.64[c]	34.33±2.78[bc]	34.77±2.76[ba]	34.59±0.43

Means followed by the same letter in the same row are not significantly different according to Duncan s multiple range (α=0.05).

smaller than Carniolan and Italian honey bees. This could be attributed mainly to the uncontrolled hybridisation among managed colonies of El-Behera due to migratory beekeeping activity, especially since the colonies are distributed in relatively small geographical areas. It is known that hybrids of Egyptain X Carniolan honey bees are common throughout Egypt, due to the importation of large numbers of Carniolan honey bees mainly between 1930 to 1940 (Sheppard *et al.*, 2001). Other honey bee subspecies were also imported to Egypt (Page *et al.,* 1981), mainly Italian honey bees. This explains why the measured characteristics are larger than those of native bees and smaller than Carniolan or Italian honey bees.

Track changes of measured characteristics

The current bees of El-Behera are slightly larger than in the past (2007) and only few decreases happened in some characteristics. Unlike the results obtained by Abou-Shaara *et al.* (2012b), that found that most of measured characteristics of El-Behera districts showed a decrease in 2007 compared to 2006. The results of this study prove that the reduction in morphological characteristics of El-Behera honey bees was temporary. Various studies highlighted the impacts of environmental conditions (Milne *et al.*, 1986), hybridisation with other honey bee subspecies (Garnery *et al.*, 1998) and migratory beekeeping (Marghitas *et al.*, 2008) on morphology of honey bees. Thus,
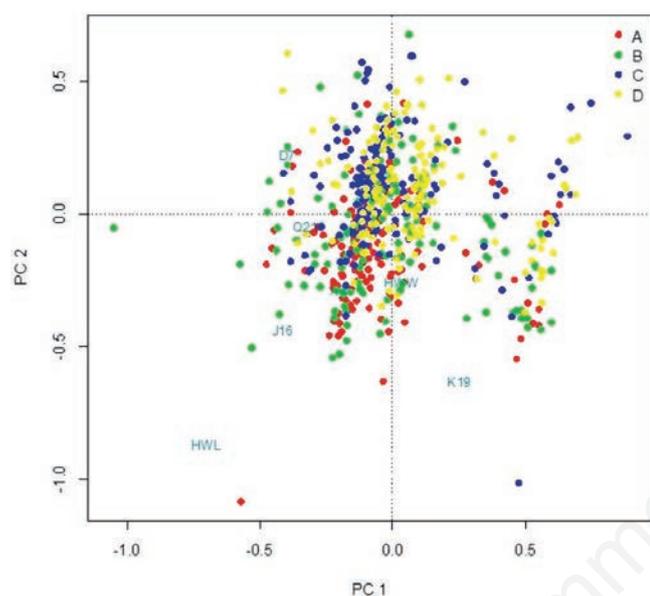
Figure 3. Principal component analysis results for honey bee colonies of studied districts (A, Damanhour; B, El-Mahmoudia; C, El-Bostan; D, Hosh Esa).
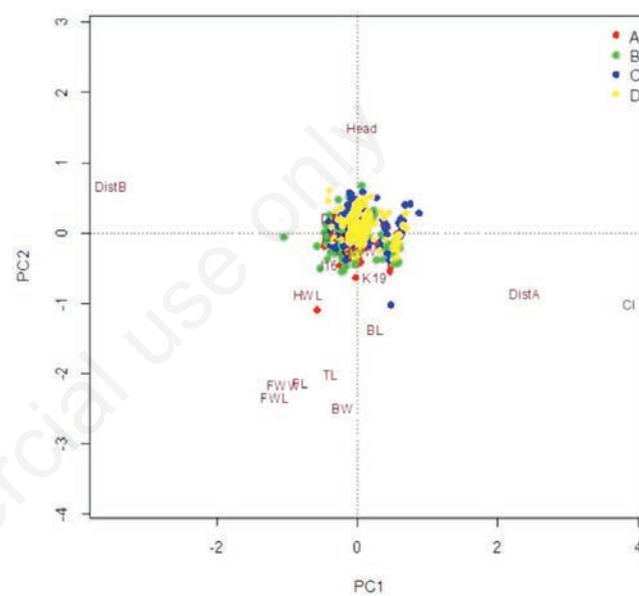
Figure 4. Variables used in principal component analysis for honey bee colonies of studied districts (A, Damanhour; B, El-Mahmoudia; C, El-Bostan; D, Hosh Esa) and studied characteristics. FWL, fore wing length; FWW, fore wing width; HWL, hind wing length; HWW, hind wing widt; FL, femur length; TL, tibia length; BL, basetarsus length; BW, basetarsus width; DistA, distance A; DistB, distance B; CI, cubital index.

Table 2. Differences between means of morphological characteristics for studied districts during 2007 and 2014. The values are in mm except for cubital index, and the values were calculated as (2007 means - 2014 means).

Characteristics	Districts	Damanhour	El-Mahmoudia	El-Dalangat Hosh
Fore wing length	0.03	0.02	0.05	0.09
Fore wing width	−0.08	−0.18	−0.06	−0.12
Hind wing length	−0.04	0.02	0.01	0.1
Hind wing width	0.01	0.01	−0.01	0.01
Femur length	−0.10	−0.10	−0.04	−0.03
Tibia length	−0.16	−0.12	−0.12	−0.03
Basetarsus length	0.06	0.12	0.14	0.05
Basetarsus width	−0.08	0.00	0.02	0.01
Cubital index	0.40	−0.15	−0.50	−0.54
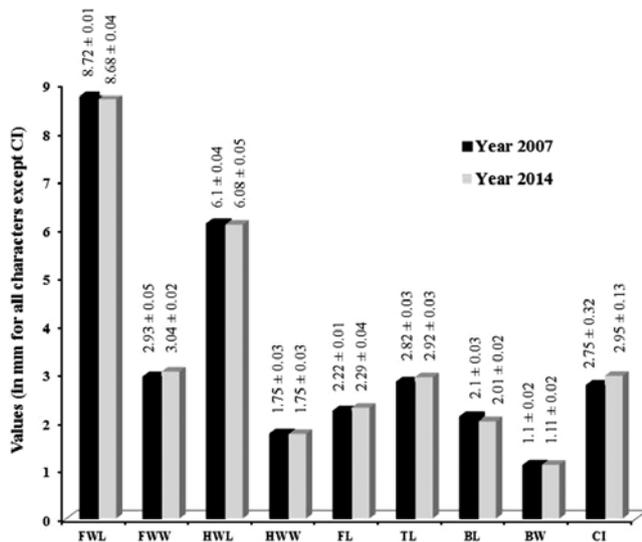
Figure 5. Overall means for 2007 and 2014. FWL, fore wing length; FWW, fore wing width; HWL, hind wing length; HWW, hind wing widt; FL, femur length; TL, tibia length; BL, basetarsus length; BW, basetarsus width; CI, cubital index.

Figure 6. Comparison between different years. Principal component analysis for honey bee colonies of studied districts (A, Damanhour; B, El-Mahmoudia; C, El-Bostan; D, Hosh Esa) during 2007 and 2014. FWL, fore wing length; FWW, fore wing width; HWL, hind wing length; HWW, hind wing widt; FL, femur length; TL, tibia length; BL, basetarsus length; BW, basetarsus width; CI, cubital index.

the uncontrolled hybridisation between managed honey bee colonies due to beekeeper practices including migratory beekeeping could be the main reason behind the slight increase in morphological characteristics. Also, it may be that new hybrid queens were recently introduced to El-Behera and widely used. The previous investigations by Kolmes & Sam (1991), Edriss *et al*. (2002), and Mostajeran *et al*. (2006) have highlighted the positive correlation between morphological characteristics and colony productivity. Thus, it could say that colonies productively have not impacted since 2007. It could be expected that the current bees with slightly larger characteristics than the past are good in regard to colony productivity.

Conclusions

Current honey bees of El-Behera are slightly larger than those of 2007. High degree of hybridisation in studied colonies was found. No major changes have happened to current bees since 2007, and colonies productivity is expected to be the same as 2007. Measuring the morphological characteristics in different years is advisable to follow the changes in time that have occurred to honey bees.

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