

Allometric studies on *Apis mellifera adansonii* workers from three ecological zones of Nigeria

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Abstract

This study evaluated the body allometry of honey bees, *Apis mellifera adansonii* Latreille (Hymenoptera Apidae) from the rainforest, Guinea savannah and derived savannah zones of Nigeria. Fifteen honey bee workers were collected from fifty-four colonies comprising of eighteen colonies in each of the three ecological zones. Honey bee samples were dissected and morphological parameters measured using PhotoScan method. The relationship between the body parts and total body length was calculated using linear regression. Length-weight relationship was analysed by using the allometric linear relationship. Results showed significantly higher ($P < 0.05$) mean body weight, total body length and mouth width in the honey bees from the rainforest zone. The thorax and abdomen lengths of honey bees from the rainforest and Guinea savannah zones showed significant linear relationship with the total body length of the honey bees. Also, lengths of abdomen, thorax, head, antenna, proboscis, pollen basket, maxillae, hind limb, fore wing and hind wing of honey bees from the three ecological zones exhibited negative allometric growth pattern with the total body length. Length-weight relationship however revealed a positive allometric growth pattern in honey bees from the rainforest zone and negative allometric in those from Guinea savannah and derived savannah zones. Thus, length-weight relationship could be a more sensitive tool in assessing the allometric pattern of honey bees from different environments.

Key words: honey bees, growth pattern, morphometry, environment, adaptation.

Introduction

Beekeeping, being a branch of agriculture called apiculture involves the rearing and management of honey bees in hives for the production of valuable materials such as honey, beeswax, propolis, honey bee pollen, honey bee venom and royal jelly (Ojeleye, 1999; Shuaib *et al.*, 2009). Beekeeping has long been practiced as occupation in Nigeria (Olagunju, 2000). This act of beekeeping has been recorded in different states of Nigeria. Such include Ogun, Oyo, Osun, Ondo, Lagos and Ekiti states (Matanmi *et al.*, 2008; Lawal and Banjo, 2010; Fakayode *et al.*, 2010; Ezekiel *et al.*, 2013); Adamawa state (Abdullahi *et al.*, 2011); Abia state (Onwumere *et al.*, 2012); Kwara state (Ajao *et al.*, 2014); Benin, Delta, Bayelsa, Cross River, Rivers and Akwa-Ibom states (Adenekan *et al.*, 2014); Enugu state (Onyekuru *et al.*, 2010); Kaduna and Abuja (Ige and Modupe, 2010) and Edo state (Folayan and Bifarin, 2013).

Morphological structures are the most noticeable aspect of the phenotype of an organism and are usually an interface between the genotype and the organism's environment (Francoy *et al.*, 2006). Morphometrics, according to Oyerinde (2017), was described as the measurement and analysis of morphological characters that could be widely used to study the systematics, life history and physiology of insects. Morphometrics could also be used to measure both phenotypic and genotypic characteristics. These characteristics was described by Kekeçoğlu *et al.* (2007) to include body size, pigmentation, and venation of fore wing in *Apis mellifera* L. (Hymenoptera Apidae). Previous studies have used morphometric analysis as a tool in the assessment of subspecies limits in *A. mellifera* (Ruttner, 1988), and in the evaluation of genetic variability in honey bees (Kekeçoğlu *et al.*, 2007).

The term allometry explains the change of body characteristics of living beings with size and the scaling relationship between the size of a part of the body and the whole body size (Shingleton, 2010). Report has shown that allometry has been viewed on three different perspectives such as static allometry, phylogenetic or evolutionary allometry and ontogenetic allometry (Klingenberg and Zimmermann, 1992). Static allometry was described as the scaling relationship between body organs and total body size at a single developmental stage or after growth cessation (Stern and Emlen, 1999). Ontogenetic allometry explains the growth path of a body organ in relation to body size during growth while evolutionary allometry is the size relationship between organs across species (Stern and Emlen, 1999). Reports have also identified allometry as a standard quantitative method for the evolutionary study of variations in size relationships between different body parts (Vencl, 2004) in several animal groups, vertebrates and invertebrates (Calabuig *et al.*, 2013; Pelabon *et al.*, 2014; Stern and Emlen, 1999). Shingleton *et al.* (2010) suggested a high correlation between static and ontogenetic allometry in insects because the patterns of static allometry of insects are established in the period of larval development. Therefore, changes in morphological pattern in relation to the different types of allometry could be better used to explain the morphological differentiation commonly observed in insects with respect to species or evolutionary lineage, caste, age, population and sex (Paciencia *et al.*, 2012).

In their report, Owen and Harder (1995) suggested that if honey bee colonies differ in allometric relations, differential productivity may occur among the colonies. There is however a dearth of information on the allometry of the Nigerian honey bee populations. Similarly, the Nigeria landmass consists of a variety of ecological zones ranging

from the rainforest to Sahel savannah zone. While allometry equations have been constructed for various social insects, little attention has been given to the comparison between colonies (Owen and Harder, 1995). Thus, it is essential to study the allometric relationships of different colonies of the honey bees from the different ecological zones of the Nigeria using their morphological parameters. This study therefore evaluated the body allometric relationship of the honey bees, *Apis mellifera adansonii* Latreille (Hymenoptera Apidae) from the rainforest, Guinea savannah and derived savannah zones of Nigeria.

Materials and methods

Study locations

This study was conducted in the rainforest zone (Ogun state), Guinea savannah zone (Oyo state) and the derived savannah zone (Kwara state) of Nigeria. Ogun and Oyo states are located in the south-western region of the country while Kwara state is located in the middle-belt region of the country. Two apiaries from each of the ecological zones were selected and used as the study site. The study locations are described in table 1.

Sample collection

Honey bee workers of unknown age were collected by modifying the method of Ajao and Babatunde (2013). The honey bee hives were gently tapped from the side. As soon as the honey bees began to rush out of the hive, a perforated plastic jar was used to trap the honey bees from the entrance of the hive. Where the entrance is not obvious, the jar was swirled to collect the bees. Collected honey bee samples were transported in ice box. Samples of collected honey bees were identified in the Biology laboratory of Kwara state University, Malete, Kwara state, Nigeria.

Body weight

The body weights of the honey bees were measured in the laboratory using an electric scale (MH-Series) with calibration of 0.01 g to 200 g.

Morphometric analysis

A total of 810 honey bee workers were collected from the study locations for morphometric studies. These were

preserved in formol-alcohol solution containing 70% ethanol, 25% water and 5% formalin as described by Carreck *et al.* (2013). The honey bee samples were dissected to dismember body parts following the standard method of *A. mellifera* dissection described by Carreck *et al.* (2013).

The morphometric parameters examined include:

- Total body length;
- Head length and head width;
- Thorax length;
- Abdomen length;
- Length of antenna;
- Mouth parts - length of proboscis, maxillae and labial palp, mouth width;
- Hind limb lengths - femur, pollen basket length and width, basitarsus and tarsi length;
- Fore wing and hind wing length and width
- Length of the radial cell.

The morphometric measurements were done by modifying the method described by Kamel *et al.* (2013). The method integrates a photo scanner and Photoshop program, called the Scan Photo Method. However, this present study used a digital microscope (GLChina - 0.3 CMOS Sensor) instead of a photo scanner. The digital microscope was connected to a computer system to take the picture of the honey bee parts for measurement. The morphometric measurements of the pictured parts were done on-screen using Adobe Photoshop CS6 Extended. The software was calibrated with the digital microscope using a stage micrometer. The calibration was re-affirmed at each measurement. Pictures of some of the measured honey bee parts are also shown in figure 1.

Identification of common flora

Common plant species around the study areas were collected for identification. Some of the plant species, especially the trees were identified on-site. Other plant species were collected and identified in the Biology Laboratory, Kwara State University, Malete Kwara State Nigeria.

Statistical analyses

Data obtained were subjected to statistical analyses using SPSS (Statistical Package for Social Sciences) version 20.0 (IBM, 2011). Analysis of variance was used to compare means. Mean result values were presented as

Table 1. Description of the study locations.

States	Rainforest		Guinea savannah		Derived savannah	
	Ogun		Oyo		Kwara	
Apiary locations	1. Agbungburu village, Osiele	2. Omiseeni village, Odeda	1. Tesi Garba village, Igbeti	2. Aba-Ogbomoso, Saki	1. Malete-KWASU-CBTR	2. Buari-BTRC
Coordinates	3°31'13.6"E 7°11'11.6"N	3°30'05.5"E 7°13'07.6"N	4°02'34.1"E 8°51'56.1"N	3°25'10.4"E 8°41'47.3"N	4°28'34.4"E 8°42'57.5"N	4°56'13.8"E 8°28'39.8"N
Rainfall	1000 to 2000 mm		800 to 1800 mm		1154 to 1885 mm	
Temperature	24.4 °C to 28.5 °C		25 °C to 28.8 °C		26.2 °C to 30.6 °C	
Altitude	148 to 175 m a.s.l.		365 to 427 m a.s.l		333 to 422 m a.s.l	
Type of hive	Kenya top bar		Kenya top bar		Kenya top bar	

KWASU-CBTR - Kwara State University Centre for Beekeeping Training and Research, Malete; BTRC - Beekeeping Training and Research Centre, Buhari.

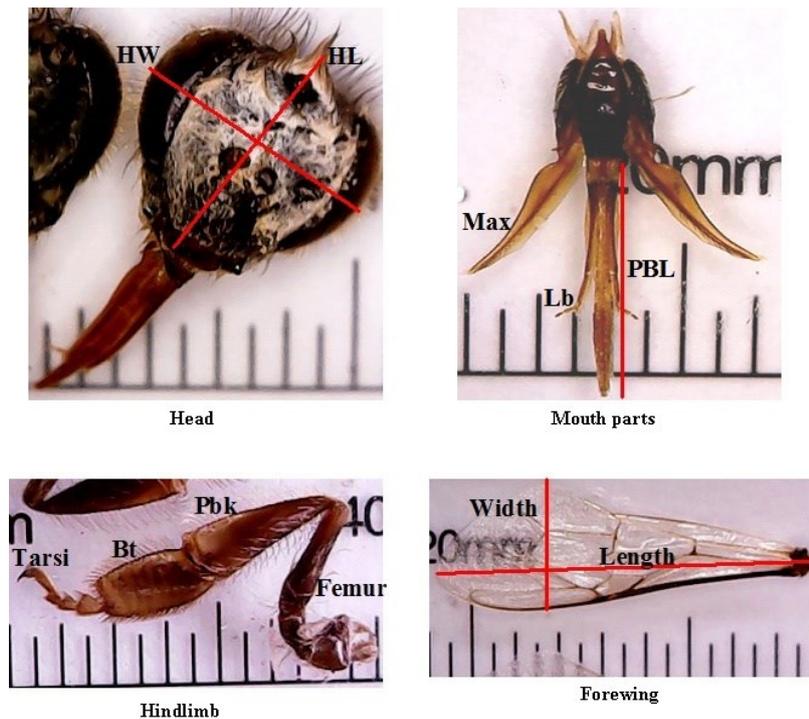


Figure 1. Morphometrics of the head, mouthparts, hind limb, antenna, fore wing and hind wing of the honey bee; HW = head width; HL = head length; Max = maxillae; Lb = labial palp; PBL = proboscis length; Bt = basitarsus; Pbk = pollen basket.

mean \pm standard deviation. Post hoc test used was the Student-Newman-Keuls. Statistical significance was based on $P < 0.05$. The relationship between the body parts and total body length was determined using linear regression. The length-weight relationship was analysed by using the allometric linear relationship: $\text{Log}(W) = \text{Log}(a) + b \times \text{Log}(L)$. Where W = weight of honey bees in gram; L = length of honey bees in mm; a = describe the rate of change of weight with length (allometric intercept); and b = weight at unit length (allometric slope) (Pelabon *et al.*, 2014). The equation was log transformed to estimate the parameters 'a' and 'b'. Growth pattern was predicted to show negative allometry when b is less than 1 ($b < 1$) and positive allometry when b is greater than one ($b > 1$) (Pelabon *et al.*, 2014).

Results

Morphometric structures

Mean body weight of the honey bees was significantly ($P = 0.001$) different between the ecological zones (table 2). This ranged from 0.053 ± 0.01 g in the derived savannah zone to 0.059 ± 0.02 g in the Guinea savannah zone and 0.064 ± 0.02 g in the rainforest zone. Similarly, the total body length and mouth width were significantly highest in the honey bees from the rainforest zone. These were not significantly ($P > 0.05$) different between the Guinea savannah and the derived savannah zones. On the other hand, honey bees from the derived savannah zone had significantly higher head region length and the lowest abdomen length. There was however no significant difference recorded in the mean thorax length, head length,

head width, antenna length, proboscis length, maxillae length, labial palp length, wings (fore wing and hind wing) length and width, radial cell length, total hind limb length, femur length, pollen basket length and basitarsus length among the honey bees from the three ecological zones.

Body allometry of the honey bees

Results also showed that the abdomen, thorax, head, antenna, proboscis, pollen basket, maxillae, hind limb, fore wing and hind wing of bees from the rainforest, Guinea savannah and the derived savannah zones of Nigeria exhibit negative allometric growth pattern (figures 2, 3 and 4). The thorax and abdomen length of bees from the rainforest and Guinea savannah zones showed significant ($P < 0.05$) linear relationship with the total body length of the bees (figure 2). This relationship was not significant among the bees from the derived savannah zone. Similarly, proboscis, pollen basket and hind wing lengths only showed significant relationship with the total body length among the bees from the rainforest zone (figures 3 and 4).

Length of proboscis, lengths of the bee mouth parts

The linear relationship between the length of proboscis and lengths of the bee mouth parts is represented in figure 5. The length of pollen basket was significantly related to the length of proboscis ($r = 0.164$, $P = 0.01$) and the length of the maxillae ($r = 0.122$, $P = 0.11$). These relationships were however weak and positive. On the other hand, the relationship between the length of pollen basket and the length of labial palp was very weak and not significant ($r = 0.055$, $P = 0.23$).

Table 2. Morphometric parameters of honey bees (means \pm standard deviation) from rainforest, derived savannah and Guinea savannah zones of Nigeria. Means followed by the same letter, within the rows, are not significantly different.

	Rainforest	Guinea savannah	Derived savannah
Total body weight (g)	0.064 \pm 0.02 ^a	0.059 \pm 0.02 ^b	0.053 \pm 0.01 ^c
Total body length (mm)	9.56 \pm 0.44 ^a	9.27 \pm 0.37 ^b	9.30 \pm 0.43 ^b
Head region (mm)	1.58 \pm 0.47 ^b	1.47 \pm 0.52 ^b	1.81 \pm 0.64 ^a
Thorax length (mm)	3.43 \pm 0.24 ^a	3.35 \pm 0.20 ^a	3.32 \pm 0.24 ^a
Abdomen length (mm)	4.54 \pm 0.54 ^a	4.44 \pm 0.45 ^a	4.16 \pm 0.40 ^b
Head length (mm)	3.13 \pm 0.07 ^a	3.14 \pm 0.08 ^a	3.18 \pm 0.17 ^a
Head width (mm)	3.70 \pm 0.11 ^a	3.69 \pm 0.09 ^a	3.69 \pm 0.16 ^a
Mouth width (mm)	1.80 \pm 0.46 ^a	1.68 \pm 0.09 ^b	1.67 \pm 0.20 ^b
Antenna length (mm)	3.94 \pm 0.11 ^a	3.93 \pm 0.11 ^a	3.97 \pm 0.13 ^a
Proboscis length (mm)	5.19 \pm 0.30 ^a	5.21 \pm 0.31 ^a	5.28 \pm 0.27 ^a
Maxillae length (mm)	3.83 \pm 0.10 ^a	3.81 \pm 0.16 ^a	3.83 \pm 0.16 ^a
Labial palp length (mm)	2.61 \pm 0.10 ^a	2.62 \pm 0.11 ^a	2.63 \pm 0.11 ^a
Fore wing length (mm)	8.47 \pm 0.26 ^a	8.36 \pm 0.19 ^a	8.45 \pm 0.25 ^a
Fore wing width (mm)	2.92 \pm 0.15 ^a	2.94 \pm 0.10 ^a	2.95 \pm 0.11 ^a
Hind wing length (mm)	5.65 \pm 0.33 ^a	5.55 \pm 0.17 ^a	5.60 \pm 0.20 ^a
Hind wing width (mm)	1.70 \pm 0.06 ^a	1.70 \pm 0.08 ^a	1.72 \pm 0.08 ^a
Radial cell length (mm)	2.93 \pm 0.08 ^a	2.95 \pm 0.09 ^a	2.96 \pm 0.08 ^a
Total hind limb length (mm)	9.06 \pm 0.37 ^a	9.02 \pm 0.45 ^a	9.15 \pm 0.30 ^a
Femur length (mm)	2.31 \pm 0.16 ^a	2.24 \pm 0.14 ^a	2.30 \pm 0.13 ^a
Pollen basket length (mm)	2.93 \pm 0.30 ^a	2.86 \pm 0.38 ^a	2.99 \pm 0.16 ^a
Basitarsus length (mm)	2.06 \pm 0.15 ^a	2.07 \pm 0.13 ^a	2.09 \pm 0.11 ^a

Length-weight relationship of the honey bees

The length-weight relationship of the honey bees was significant ($P = 0.01$) with relatively weak coefficient of determination ($R^2 = 0.130$) among the bees from the rainforest zone (table 3). On the other hand, the length-weight relationship recorded among the bees from the Guinea savannah and the derived savannah zones were not significant ($P > 0.05$). The growth pattern of the bees as determined from the length-weight relationship was positive allometric among the bees from the rainforest zone while those from the Guinea savannah and the de-

derived savannah zones had negative allometric growth pattern.

On the overall, honey bees from the study area (combined data from the rainforest, Guinea savannah and the derived savannah zones of Nigeria) showed significant length-weight relationship ($P = 0.01$) with mean body length of 9.35 ± 0.02 mm and body weight of 0.059 ± 0.02 g and a positive allometric growth pattern. The graphical representation of the length-weight relationship of the bees from each of the ecological zones and the overall study area is also represented in [figure 6](#).

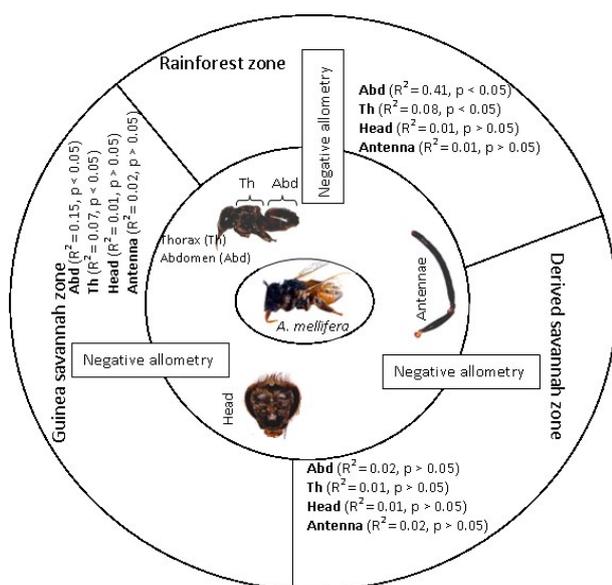


Figure 2. Allometric pattern of the head, antenna, thorax and abdomen lengths of *A. mellifera* in three ecological zones of Nigeria.

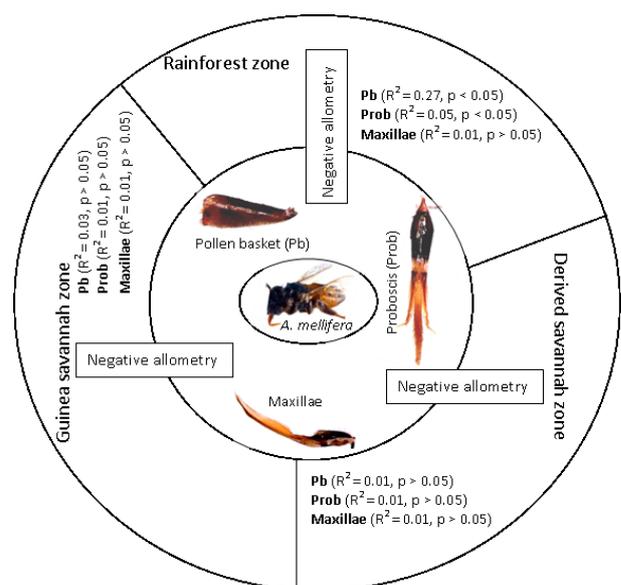


Figure 3. Allometric pattern of the foraging structures (proboscis, maxillae and pollen basket) of *A. mellifera* in three ecological zones of Nigeria.

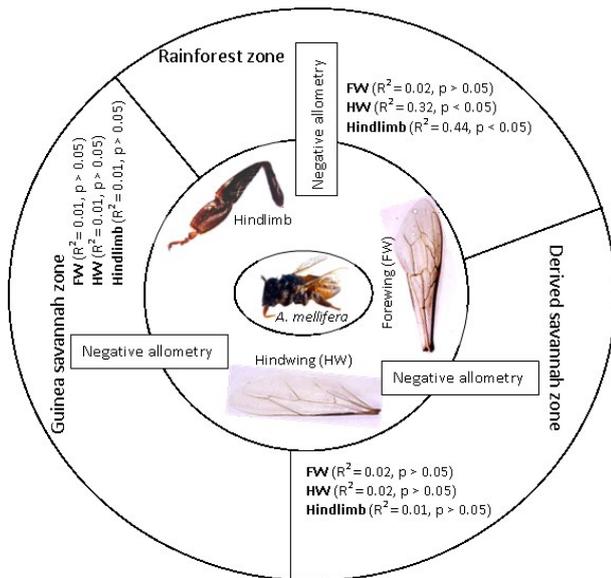


Figure 4. Allometric pattern of the wings and hind limb lengths of *A. mellifera* in three ecological zones of Nigeria.

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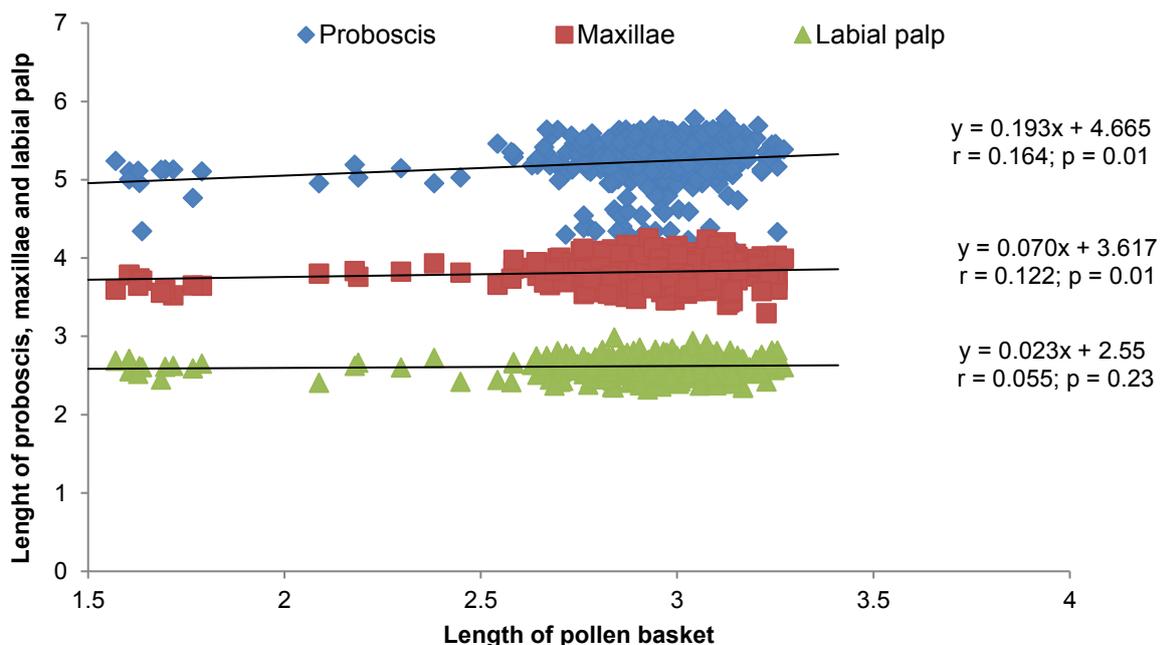


Figure 5. Linear relationship between the length of proboscis and lengths of the bee mouth parts.

Table 3. Length-weight relationship and growth pattern of the bees from the three ecological zones and the entire study area. Means \pm standard deviation.

	Rainforest	Guinea savannah	Derived savannah	Overall study area
Total length (mm)	9.56 \pm 0.44	9.27 \pm 0.37	9.30 \pm 0.43	9.35 \pm 0.02
Weight (g)	0.064 \pm 0.02	0.059 \pm 0.02	0.053 \pm 0.01	0.059 \pm 0.02
Intercept (a)	1.649	-2.19	-1.762	-2.232
Slope (b)	2.797	0.942	0.494	1.025
Correlation coefficient (r)	0.360	0.175	0.151	0.178
Coefficient of determination (R^2)	0.130	0.031	0.023	0.032
P value	0.01	0.12	0.18	0.01
Growth pattern	Positive allometric	Negative allometric	Negative allometric	Positive allometric
Exponential equation	Wt = 1.649(TL) ^{2.797}	Wt = 2.190(TL) ^{0.942}	Wt = 1.762(TL) ^{0.494}	Wt = 2.232(TL) ^{1.025}

Wt = weight; TL = total length.

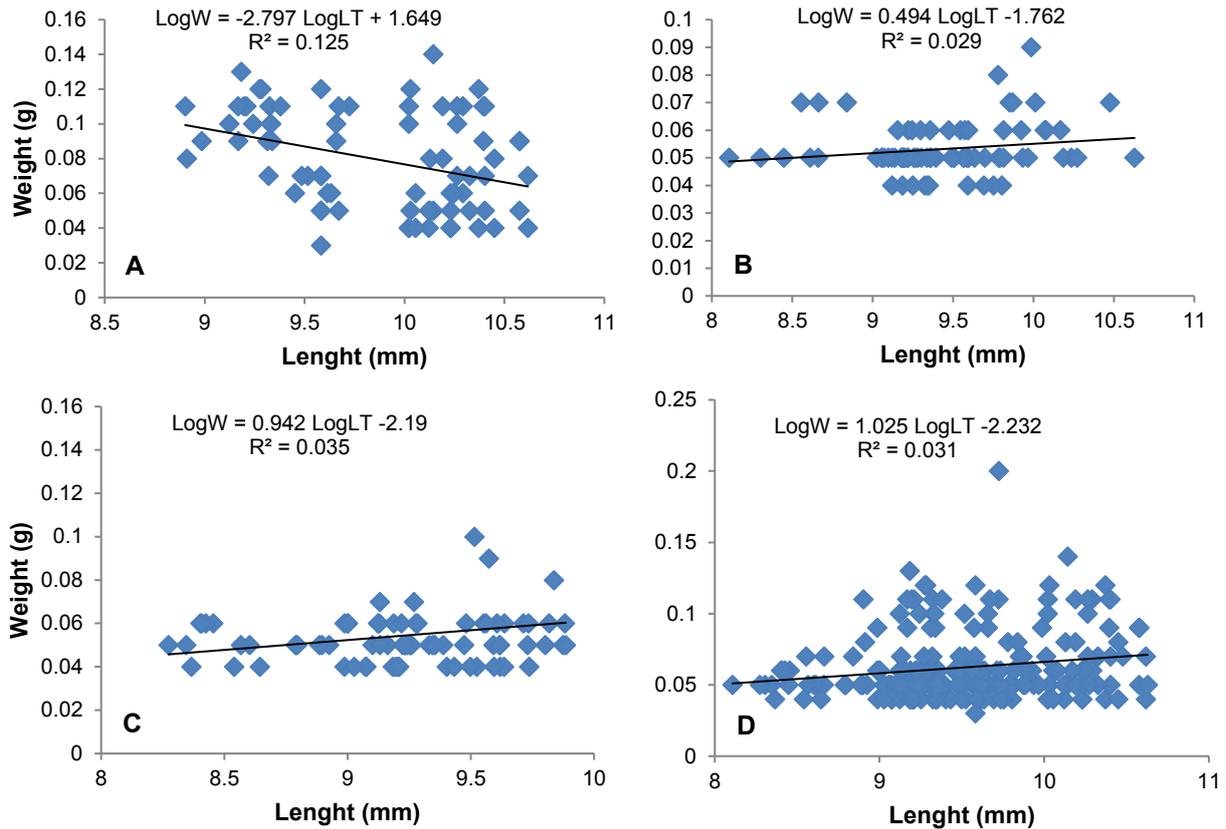


Figure 6. Length-weight relationships of the bees: A) Rainforest zone; B) Derived savannah zone; C) Guinea savannah zone; D) Overall study area. W = Weight; TL = Total length.

Diversity of bee flora around the study areas

A total of twenty bee flora were identified in the study apiaries of the rainforest, Guinea savannah and derived savannah zones (table 4). Flora diversity was higher in the Guinea savannah zone (13 species) than the rainforest

and the derived savannah zones (10 species each). Of the entire common flora identified, *Anacardium occidentale*, *Azadiracta indica*, *Mangifera indica* and *Tridax procumbens* were present in the three ecological zones.

Table 4. Common flora identified around the study areas (+ present; – absent).

Plant species	Common name	Rainforest	Guinea savannah	Derived savannah
<i>Acacia nilotica</i>	Acacia	+	+	–
<i>Ageratum conyzoides</i>	Goat weed	–	–	+
<i>Amaranthus</i> spp.	Pigweed	–	–	–
<i>Anacardium occidentale</i>	Cashew	+	+	+
<i>Ananas cosmosus</i>	Pineapple	+	–	–
<i>Azadiracta indica</i>	Neem	+	+	+
<i>Citrus</i> sp.	Oranges	–	–	+
<i>Cynodon dactylon</i>	Bahama grass	+	+	–
<i>Delonix regia</i>	Flame of forest	–	+	–
<i>Gmelina arborea</i>	Gmelina tree	+	–	–
<i>Gossypium hirsutum</i>	Cotton	–	+	–
<i>Hibiscus sabradifa</i>	Roselle	–	+	–
<i>Glycine max</i>	Soybeans	–	–	+
<i>Mangifera indica</i>	Mango	+	+	+
<i>Mimosa pudica</i>	Sensitive plant	+	–	+
<i>Moringa oleifera</i>	Drumstick tree	–	+	–
<i>Parkia biglobosa</i>	Locust bean tree	–	+	+
<i>Tectona grandis</i>	Teak	+	–	+
<i>Tridax procumbens</i>	Coat button	+	+	+
<i>Vitellaria paradoxa</i>	Shea tree	–	+	–

Discussion

This study evaluated the allometry of honey bees from the rainforest, Guinea savannah and derived savannah zones of Nigeria. Higher body weight was recorded in the honey bees and this could be associated to the length of foraging time. The savannah is the central biome in the transition between grasslands and forests with the characteristics of trees and grasses coexistence (Baudena *et al.*, 2015). Thus, the rainforest zone is naturally endowed with greater diversity of honey bee flora which is denser in population than the Guinea savannah and the derived savannah zones respectively. The denser population of forage plants in the rainforest zone could have resulted in shorter foraging length in the honey bees from the rainforest zone, hence higher body weight. Similarly, significantly higher total body length and mouth width were recorded in the honey bees from the rainforest zone. Previous study has associated the increase in sizes of the external morphologic characters of honey bees to food abundance (Ajao *et al.*, 2014). Other studies also affirmed that environmental changes have a direct influence on honey bee growth and development (Kandemir *et al.*, 2005; Mladenovic *et al.*, 2011). Therefore, Abou-shaara *et al.* (2012) submitted that the morphologic characters and weight are reflections of the strong influence of the environment in the morphology of *A. mellifera*. Hence, it is unexpected that the honey bees from the savannah zones would have lesser weight and body length as a result of used energy during forage from the disperse and less denser plant population than those from the rainforest zone.

This study has revealed that the body structures of the honey bees from the three ecological zones of Nigeria exhibited negative allometry with the total body length. During allometry studies, a negative allometry shows that the size of a structure grows at a shorter rate than the generalized body size (Monteiro, 1997). The implication of this is that the body parts of honey bee workers from the studied ecological zones of Nigeria grows in length at a slower rate than the total body length. Some of the evaluated body parts include the head, antenna, proboscis, wings, pollen basket and the total hind limb. These structures contributes greatly to the foraging success of the honey bees. For instance, the head serves as the major sensory region of the bees' body with two compound eyes and three simple eyes located on the top of the head (Suwannapong *et al.*, 2011). Reports have also shown that the antennae is associated with asymmetry of anatomy (i.e. sensilla and electroantennographic antennal neurons) (Rogers and Vallortigara, 2008; Anfora *et al.*, 2010) capable of detecting odour (Letzkus *et al.*, 2006). Also, the importance of the proboscis to insects with sucking mouth parts has been reported to include uptake of surface liquids or nectar from flowers (Kingsolver and Daniel, 1995). It is therefore possible that the growth of these important forage structures in the honey bees is dependent more on their usage during foraging.

On the other hand, length-weight relationship of honey bees from the three ecological zones showed varied growth pattern. For example, honey bees from the rainforest zone exhibited positive allometry while those from the Guinea savannah and derived savannah zones exhibited negative allometry. According to Monteiro (1997),

positive allometry shows that a given structure grows at a higher rate than generalized body size while a negative allometry indicates that the structure grows at a shorter rate. Thus, it could be deduced that the honey bees from the rainforest zones grows faster in weight than the total body length and the other way round in the Guinea savannah and derived savannah zones. This is in consonant with the result of this study which recorded significantly higher mean body weight recorded in the honey bees from the rainforest zone than those from the Guinea savannah and derived savannah zones. The report of Owen and Harder (1995) have earlier suggested that geographic variation could affect the allometry of the bumblebee.

Conclusion

This study has shown that the allometric pattern of the body parts of honey bees in relation to the total body length is not significantly influenced by the ecological zones. However, allometric pattern of the honey bees measured using the length-weight relationship was significantly influenced by the ecological zones. Thus, length-weight relationship could be a more sensitive tool in assessing the allometric pattern of honey bees from different environments.

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