UNIVERSIDADE TECNOLÓGICA FEDERAL DO PARANÁ
PROGRAMA DE PÓS-GRADUAÇÃO EM ZOOTECNIA

PATRICK ROMUALD FOTSO KENMOGNE

GENETIC ASPECTS OF MORPHOMETRIC TRAITS AND REPRODUCTIVE ORGANS OF AFRICANIZED HONEY BEE DRONES, *Apis mellifera L.* (HYMENOPTERA: APIDAE)

DISSERTAÇÃO

DOIS VIZINHOS
2018

PATRICK ROMUALD FOTSO KENMOGNE

GENETIC ASPECTS OF MORPHOMETRIC TRAITS AND REPRODUCTIVE ORGANS OF AFRICANIZED HONEY BEE DRONES, *Apis mellifera L.* (HYMENOPTERA: APIDAE)

Dissertação apresentada ao Programa de Pós-Graduação em Zootecnia, da Universidade Tecnológica Federal do Paraná, Câmpus Dois Vizinhos, como requisito parcial para obtenção do Título de “Mestre em Zootecnia” - Área de Concentração: Produção Animal

Orientadora: Profa. Dra. Fabiana Martins Costa Maia

DOIS VIZINHOS

2018

F759g Fotso Kenmogne, Patrick Romuald.
Genetic aspects of morphometric traits and reproductive organs of Africanized honey bee drones, *Apis mellifera L*. (Hymenoptera: Apidae). / Patrick Romuald Fotso Kenmogne – Dois Vizinhos, 2018.
45 f.

Orientadora: Dra. Fabiana Martins Costa Maia.
Dissertação (Mestrado) - Universidade Tecnológica Federal do Paraná, Programa de Pós-Graduação em Zootecnia, Dois Vizinhos, 2018.
Bibliografia p.42-45.

1. Abelhas africanizadas. 2. Abelhas africanizadas - Aspectos genéticos. 3. Órgãos reprodutores. I. Maia, Fabiana Martins Costa, orient. II. Universidade Tecnológica Federal do Paraná – Dois Vizinhos. III. Título

CDD: 595.799

Ficha catalográfica elaborada por Keli Rodrigues do Amaral Benin CRB: 9/1559

Biblioteca da UTFPR-Dois Vizinhos

Ministério da Educação
Universidade Tecnológica Federal do Paraná
Câmpus Dois Vizinhos
Diretoria de Pesquisa e Pós-Graduação
Programa de Pós-Graduação em Zootecnia

UNIVERSIDADE TECNOLÓGICA FEDERAL DO PARANÁ

TERMO DE APROVAÇÃO

Título da Dissertação n° 103

Genetic aspects of morphometric traits and reproductive organs of Africanized honey bee drones, *Apis mellifera L.* (Hymenoptera: Apidae)

Patrick Romuald Fotso Kenmogne

Dissertação apresentada às nove horas do dia dez de julho de dois mil e dezoito, como requisito parcial para obtenção do título de MESTRE EM ZOOTECNIA, Linha de Pesquisa – Produção e Nutrição Animal, Programa de Pós-Graduação em Zootecnia (Área de Concentração: Produção animal), Universidade Tecnológica Federal do Paraná, Câmpus Dois Vizinhos. O candidato foi arguido pela Banca Examinadora composta pelos professores abaixo assinados. Após deliberação, a Banca Examinadora considerou o trabalho .....

Banca examinadora:

Dr. Wagner Paris
UTFPR - DV

Dra. Juliana Morini Kupper Cardoso Perseguini
UTFPR - DV

Dra. Sandra Maria Simonelli
UEL

Coordenador do PPGZO
Assinatura e carimbo

*A Folha de Aprovação assinada encontra-se na Coordenação do Programa de Pós-Graduação em Zootecnia.

À

Deus, que permitiu que tudo pudesse ser realizado.

DEDICO...

AGRADECIMENTOS

Certamente estes parágrafos não irão atender a todas as pessoas que fizeram parte dessa importante fase de minha vida. Portanto, desde já peço desculpas àqueles que não estão presentes entre essas palavras, mas elas podem estar certas que fazem parte do meu pensamento e de minha gratidão.

À Universidade Tecnológica Federal do Paraná, Câmpus de Dois Vizinhos que por meio da instituição e de seus professores e servidores possibilitou o crescimento profissional e intelectual imprescindíveis para o desenvolvimento deste trabalho.

À professora, Dra. Fabiana Martins Costa Maia, minha orientadora, pelas horas de trabalho em conjunto, por todas as conversas, conselhos e pelo aprendizado.

À Dra. Simone Fernanda Nedel Pértile, para análise de dados.

Ao CAPES, e a Universidade Tecnológica Federal do Paraná pelo fomento com a bolsa para a realização deste Mestrado.

À todos os integrantes do grupo de pesquisa em melhoramento genético de abelhas por todo apoio e trabalho.

Aos meus amigos, Augusto Carlos Caponi de Oliveira, Bruno Lavrini e Jonathan Pia, pela amizade, parceria e momentos inesquecíveis, por me ampararem perante a todas dificuldades encontradas.

RESUMO

FOTSO KENMOGNE, Patrick Romuald. Aspectos genéticos de características morfométricas e órgãos reprodutivos de zangões Africanizados, *Apis mellifera L*. (Hymenoptera: Apidae). 2018. 45 p. dissertação (Mestrado em Zootecnia), Programa de Pós-graduação em Zootecnia, Universidade Tecnológica Federal de Panará. Dois Vizinhos, 2018.

Este estudo foi realizado no período de outubro de 2013 a abril de 2014, na Universidade Tecnológica Federal do Paraná no câmpus de Dois Vizinhos. O objetivo foi estimar os parâmetros genéticos de caracteres morfométricos, órgãos reprodutivos e a correlação genética entre caracteres morfométricos e órgãos reprodutivos de zangões africanizados maduros. Para fazer isso, modelos uni e bi característica foram usados. Dois efeitos fixos distintos foram considerados; as colmeias onde zangões foram mantidos desde a emergência até a maturidade e a época do ano. Um total de 329 zangões de 6 matrizes foram observados. Os principais resultados mostraram que os zangões apresentavam um peso corporal médio de 202,81 ± 17,84 mg. As medidas corporais médias em mm forneceram os seguintes valores: comprimento total (15,39 ± 0,74), comprimento de abdômen (7,69 ± 0,68), largura de abdômen (5,48 ± 0,29), comprimento de asa (12,40 ± 0,66) e largura de asa (3,83 ± 0,30). O peso, a área e o volume médios da vesícula seminal foram 1,80 ± 1,9 mg, 8,60 ± 2,92 mm^2 e 6,65 ± 3,31 mm^3, respectivamente. O peso, a área e o volume médios da glândula do muco foram 12,60 ± 1,9 mg, 25,45 ± 8,59 mm^2 e 37,84 ± 18,12 mm^3, respectivamente. A herdabilidade dos caracteres variou de 0,22 a 0,74. As correlações genéticas entre características morfométricas e órgãos reprodutivos variaram de -0,99 a 0,18. Todas essas características consideradas sugerem que os zangões constituem um recurso natural com variabilidade genética necessária para o melhoramento genético por ferramentas convencionais, como seleção e cruzamento.

Palavras-chave: Abelha, Correlação genética, Dimensão, Herdabilidade, Inferência bayesiana, Vesícula seminal

ABSTRACT

FOTSO KENMOGNE, Patrick Romuald. Genetic aspects of morphometric traits and reproductive organs of Africanized honey bee drones, *Apis mellifera L.* (Hymenoptera: Apidae). 2018. 45 p. Dissertation (Master's degree in Animal Science) – Postgraduate Program in Animal Science, Federal University of Technology – Paraná. Dois Vizinhos, 2018.

This study was carried out from October 2013 to April 2014 at the Federal University of Technology, Campus of Dois Vizinhos in the Paraná state of Brazil. The objective was to estimate the genetic parameters of morphometric traits, reproductive organs, and the genetic correlation between morphometric traits and reproductive organs of drones at maturity. In order to achieve this, a single-trait and two trait models were used. Two distinct fixed effects were considered; the hives where drones were kept from emergence until maturity, and the yearly time. A total of 329 drones were observed from 6 matrices. The main results showed that, honey bee drones had an average weight of 202.81 ± 17.84 mg at maturity. The average body measurements in mm gave the following values: total length (15.39 ± 0.74), length of abdomen (7.69 ± 0.68), width of abdomen (5.48 ± 0.29), length of wing (12.40 ± 0.66), and width of wing (3.83 ± 0.30). The average weight, area, and volume of seminal vesicle were respectively, 1.80 ± 1.9 mg, 8.60 ± 2.92 mm^2, and 6.65 ± 3.31 mm^3. The average weight, area, and volume of mucus gland were respectively, 12.60 ± 1.9 mg, 25.45 ± 8.59 mm^2, and 37.84 ± 18.12 mm^3. Heritability of traits ranged from 0.22 to 0.74. Genetic correlations between morphometric traits and reproductive organs ranged from -0.99 to 0.18. All these characteristics considered suggest that honey bee drones constitute a natural resource with genetic variability needed for genetic improvement by conventional tools such as selection and breeding.

Keywords: Bayesian inference, Bee, Genetic correlation, Heritability, Seminal vesicle, Size

RESUME

FOTSO KENMOGNE, Patrick Romuald. Aspects génétiques des traits morphométriques et des organes reproducteurs des faux bourdons Africanisés, *Apis mellifera L.* (Hymenoptera: Apidae). 2018. 45 p. Mémoire (Master of Science en Zootechnie), Programme de Postgraduation en Zootechnie, Université Technologique Fédérale de Panará. Dois Vizinhos, 2018.

Cette étude a été réalisée d'Octobre 2013 - Avril 2014 à l'université technologie fédérale, campus de Dois Vizinhos dans l'État de Paraná au Brésil. L'objectif était d'estimer les paramètres génétiques des traits morphométriques, des organes reproducteurs et la corrélation génétique entre les traits morphométriques et les organes reproducteurs des faux bourdons Africanisés à maturité. Pour ce faire, des modèles de un et deux traits ont été utilisés. Deux effets fixes distincts ont été considérés; les ruches où les faux bourdons ont été placés de l'émergence jusqu'à la maturité et la période de l'année. Un total de 329 faux bourdons issu de 6 matrices a été observé. Les principaux résultats ont montré que les faux bourdons avaient un poids moyen à maturité de 202,81 ± 17,84 mg. Les mesures corporelles moyennes en mm ont donné les valeurs suivantes: longueur totale (15,39 ± 0,74), longueur de l'abdomen (7,69 ± 0,68), largeur de l'abdomen (5,48 ± 0,29), longueur de l'aile (12,40 ± 0,66) et largeur de l'aile (3,83 ± 0,30). Le poids moyen, la surface et le volume de la vésicule séminale étaient respectivement de 1,80 ± 1,9 mg, 8,60 ± 2,92 mm^2 et 6,65 ± 3,31 mm^3. Le poids moyen, la surface et le volume de la glande muqueuse étaient respectivement de 12,60 ± 1,9 mg, 25,45 ± 8,59 mm^2 et 37,84 ± 18,12 mm^3. L'héritabilité des caractères variait de 0,22 à 0,74. Les corrélations génétiques entre les caractères morphométriques et les organes reproducteurs variaient de -0,99 à 0,18. Toutes ces caractéristiques considérées suggèrent que, les faux bourdons constituent une ressource naturelle avec une variabilité génétique nécessaire à l'amélioration génétique par des outils conventionnels tels que la sélection et le croisement.

Mots clés: Abeille, Corrélation génétique, Dimension, Héritabilité, Inférence bayésienne, Vésicule séminale

LISTA DAS FIGURAS

Páginas

LISTA DAS FOTOGRAFIAS

Páginas

LISTA DE TABELAS

Páginas

SUMÁRIO

Páginas

1 INTRODUCTION

The honeybee, *Apis mellífera* plays an essential role in modern agriculture. This species is responsible for providing critical ecosystem services, primarily in pollination, for a large range of high value agricultural crops (CALDERONE, 2012). The pollinator services are vital because most economies rely largely on agriculture. For example, in Brazil, the agricultural sector represents a large part of the total economy and the share of agriculture in Brazilian gross domestic product (GDP) is 11340 US$ per capita (FAO, 2014). Besides their role in pollination, honeybee make useful products such as honey and wax, as well as other nutritional, medicinal and pharmaceutical products such as royal jelly, propolis, bee venom and pollen (MICHENER, 2007). These bee products are of great economic value.

However, over the past decades, honeybee colony losses have gradually been increasing worldwide. Some researchers argue that it is necessary to implement an improved genetic evaluation methodology as a long-term solution to avoid the decline of the honeybee population (GUPTA et al., 2013). Therefore, genetic improvement by means of selective breeding requires knowledge of heritabilities of the relevant traits and of the genetic correlations between those traits (EVERT et al., 2016). Estimates of heritabilities and genetic correlations indicate the prospects for genetic improvement of traits and allow the estimation of breeding values of individuals. Subsequently, estimated breeding values can be used in breeding programmes to select genetically superior individuals to become the parents of the next generation.

Available information on honeybee breeding is scarce, especially regarding to the male caste. On the other hand, the main function of drones within the colony is to fertilize virgin queens. One of the most neglected elements of queen rearing in honeybee is the provision of suitable mates for the virgin queens (LAIDLAW; PAGE, 1998).

Studies on drones can lead to design breeding programs. Through previous study (MARTINS, 2014), it is known that the queen bee's weight at emergence is closely related to the development of its reproductive structures. According to Rhodes (2008) the unsatisfactory performance of newly mated queens is due largely to the quantity and quality of drones on mating areas. The

number of sperm and semen volume are related to the size of the drones (DUAY et al., 2002). According to Schlüns et al. (2003) there is a positive correlation between the wing size and the number of spermatozoa. Yet, most studies to date are based on phenotypic values. The estimation of heritability and genetic correlations of traits in drones can contribute to establish selection criteria**,** being more reliable than just the phenotypic value, which may underestimate the true potential of each individual genetic value.

Our objective was to estimate genetic aspects for morphometric traits and reproductive organs of Africanized honeybee drones at maturity. More specifically, it was questioned to estimate the genetic parameters of morphometric traits, reproductive organs, and the genetic correlation between morphometric traits and reproductive organs of drones at maturity, in order to set criteria for the selection of drones for reproduction.

2 Literature review

2.1 Biology of the honeybee, *Apis mellifera*

2.1.1 Origin and classification of honey bee

Honeybees probably originated in tropical Africa and spread from South Africa to Northern Europe and West into India and China. They were brought to the Americas with the first colonists and are now distributed world-wide (CULLING, 1983).

According with Michener (1974), honeybees have been classified as follows:

Kingdom: Animalia

Phylum: Arthropoda

Class: Insecta

Order: Hymenoptera

Family: Apidea

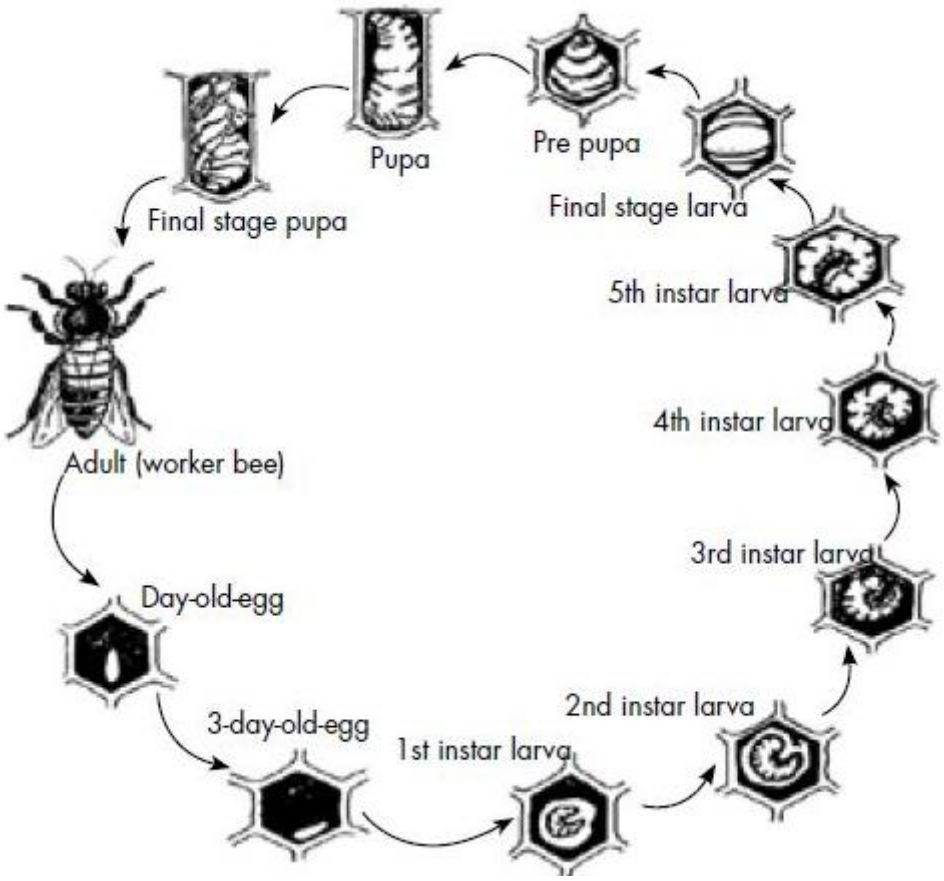
Genus: *Apis*

Species: *mellifera*

2.1.2 Life cycle and the different types of adult

Honeybees live together in a highly organized group called a colony. When bees are managed in hives, each hive houses a single colony. It is the colony that matters and tasks are accomplished through division of labour (WINSTON, 1987). Every member of the colony works not for itself, but for the benefit of the colony.

A honeybee colony has three kinds of adult: queen, worker, and drone. Each of them undergo the same four developmental stages, but the time needed to complete each stage differs. The stages are further subdivided in terms of development. The honeybee life cycle is illustrated by Figure 1.



Source: WINSTON, 1987

Figure 1: The honeybee life cycle

It is clear from Figure 1 that, the honeybee life cycle has four stages: egg, larva, pupa, and adult.

The average time taken for the different kinds of bee to complete each stage is summarized in Table 1.

Table 1: Average time taken for the different kinds of bee at each stages

	Egg stage (days)	Larval stage (days)	Pupal stage (days)	Emergence (days)
Queen	3	5	7- 8	15 - 16
Worker	3	6	11-12	20 - 21
Drone	3	7	14	24

Source: WINSTON, 1987

It stems from Table 1 that, except for the egg stage that takes 3 days, the other stages varies with the kinds of bee.

The honeybee, *Apis mellifera,* is a social insect living in a colony or a hive comprising 50000 to 80000 individuals (TCHOUMBOUÉ et al., 2010). The

population of the bee colony is composed of three different types of individuals. Table 2 shows the type, appearance, number per colony and purpose of each type of individuals.

Table 2: Type, appearance, number per colony and purpose of each types of individuals

Type	Appearance	# Per Colony	Purpose
Queen		1	Lay eggs
Worker		10,000 - 50,000	Nurse the brood; clean the colony; forage for food
Drone		100 - 500	Mate with the queens and die

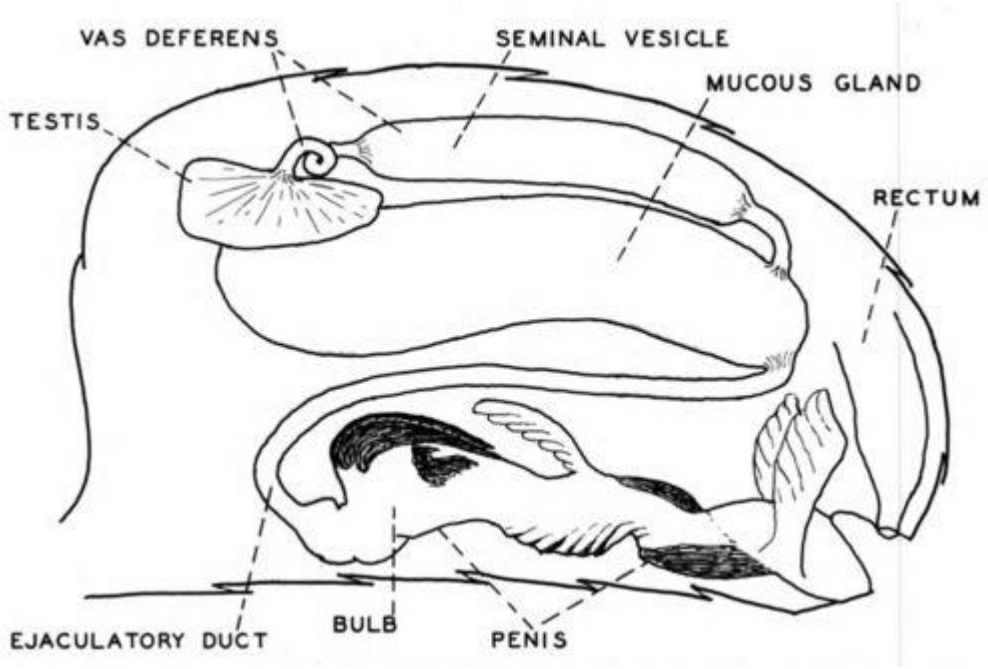
Source: TCHOUMBOUÉ et al. (2010)

As revealed by Table 2, appearance, number per colony and purpose are a function of the type of individuals.

2.1.3 Drone bee

The drones are the male bees. They do not have stings, nor do they perform hive duties. Structurally, drones are incapable of collecting food in the field. Their food supply is stopped when brood rearing slackens (ROFF, 1977). This occurs in late autumn or when a dearth period is experienced. In the hive, they are fed by worker bees. Because of limited food stores, drones are not maintained in the hive during winter. When weakened, their wings may be torn off by the workers, their legs pulled and eventually they will be dragged out of the hive. Also, drone larvae and pupae are sometimes removed from the hive. Their definite function is to mate with a virgin queen on a mating flight. When the drones are about ten days old, they are capable of impregnating a queen. This takes place outside the nest in the air (6 metres or higher) and the successful drone dies following the act. Copulation may take place up to 12.8 kilometres away from

the hive and there is considerable evidence to suggest that drones frequent drone congregating areas. A pheromone produced by the drones may identify such an area. Drones contribute to the natural equilibrium of a colony and hives from which drones have been artificially trapped and removed during principal brood rearing periods do not prosper as well as those with a relevant complement of drones. Drones that drift into a strange hive when nectar and pollen are abundant often are not attacked and are permitted to stay. The main function of drones within the colony is to fertilize virgin queens. One of the most neglected elements of queen rearing in honeybees is the provision of suitable mates for the virgin queens (LAIDLAW; PAGE, 1998). Production of viable drones is a limiting factor in successful queen rearing. Studies on drones can lead to maintain highly improved breeding programs. This can be achieved by improving the efficiency and quality of mating. Reproductive organs of drone bee is illustrated by Figure 2.



Source: OTTO et al. (1970)

Figure 2: Male reproductive organs

2.2 Genetic improvement of honeybee

The key elements to implement a breeding program are genetics parameters.

Heritability is the ratio of the variance component due to the additive effects of genes to the total phenotypic variance in a specific population. It gives an estimate of the relative importance of genetic and environmental factors. Heritability is expressed on a 0 - 1 scale, 0 implies, there are no genetic differences for that trait in the population and 1 implies, all of the observed differences are due to genetic variation. Until recently, that researchers have been estimating heritability values focused on production traits, such as kilogram of honey (MOSTAJERAN *et al.,* 2000; COSTA-MAIA, 2009; PADILHA, 2013), for gentleness (BIENEFELD & PIRCHNER, 1990) and for hygienic behaviour traits (COSTA-MAIA *et al*., 2011). However, the aforementioned traits are difficult to measure, thus the study of easy-to-measure traits associated with economical important traits is extremely relevant and that is why genetic correlations are so important.

Genetic correlation corresponds to the degree to which genes affect differences in performance for an individual trait and it is possible to estimate the extent to which different traits are affected by shared genes. Genetic correlations can be positive or negative and range from - 1 to 1. These parameters explain how pairs of traits change simultaneously. When genetic correlations are close to zero, different sets of genes control each trait and selection for one trait will have little effect on the other. Selection for one trait will increase the other if the genetic correlation is positive and decrease it if the genetic correlation is negative. A genetic correlation between traits will result in a correlated response to selection; that is the reason why genetic correlation is important in quantitative genetic and in breeding programs.

The main obstacles in the estimation of genetic parameters of colony traits in honeybees result from the fact that many characters of economic value are affected by the combined activity. To overcome these difficulties some model approaches were developed. The most advanced procedure for genetic evaluations currently available is the Best Linear Unbiased Prediction (BLUP) - animal model. BLUP has become the most widely accepted method for genetic evaluation of domestic livestock (BIENEFELD, EHRHARDT & REINHARDT, 2007). With a slight adjustment of this approach it is possible to successfully apply it to the honeybees (BIENEFELD, EHRHARDT & REINHARDT, 2007; BRASCAMP & BJIMA, 2014; RODRIGUES, 2016). BLUP, is obtained from a

linear mixed model methodology that simultaneously estimates random genetic effects. While accounting for fixed effects in the data, an optimal way and furthermore relationships among animals can be included in the model. The animal model, a linear mixed model, comprises all the relationships among all animals in the dataset.

2.3 Estimation of genetic parameters of drones, *Apis mellifera*

Estimates of genetic parameters in *Apis mellifera* bees are scarce in the world literature. For Kerr (2006), morphological, physiological or behavioral characteristics, can be improved through genetic improvement programs of *Apis mellifera* bees.

Thus, work on the additive genetic variation for the queen's weight at emergence, length and width of the abdomen, as well as their genetic associations with productive characteristics are highlighted in recent studies on Africanized queens (COSTA, 2005; HALAK, 2012; MARTINS, 2014). The parameters studied, such as heritability, allow to anticipate the possibility of success with the selection, since it reflects the proportion of the variable phenotype that can be inherited, identifying the animals that transmit these characteristics to its offspring.

2.3.1 Weight at maturity

Bigger males are considered to have a competitive advantage over smaller males when fighting for access to females (BERG et al., 1997). However, according to Schlüns et al. (2003), there is a hypothesis that the lessened reproductive success of smaller drones is caused mainly by a lower success rate in competition for access to the queen rather than reduced individual inefficiency during the copulation process. According to Rhodes (2002), honeybee colonies usually invest in larger drones but the question remains: what are the benefits of large drone production for the colony. Berg et al. (1997) stated that small drones reared in worker cells have a reproductive disadvantage compared to the normally sized drones but despite these differences, they could not identify potential proximate mechanisms for the different reproductive success.

2.3.2 Body measures

According to Schlüns et al. (2003), the wing length is an important trait since they observed a positive phenotypic correlation between it and the sperm numbers. However, in a breeding program, genetic correlations should be considered instead of phenotypic correlations, as the latter can mask, through the environmental component, the genetic component which truly indicates the animal's potential. The proportion of additive genetic variance must be estimated in the total of phenotypic variance to confirm if wing morphometric can be used as selection criterion thus aiming to obtain genetic gain in sperm production.

The study of abdomen length and width is relevant because that is where the reproductive organs are located and hence it can be used for evaluating the drone's growth and body development, conveying significant information for the identification of the populations in study. Abdomen morphometric might be useful to indicate the reproductive potential and thus can be utilized as selection criterion in order to improve the queen's and drone's reproductive performance (HALAK, 2012; MARTINS, 2014; RODRIGUES, 2016).

The drones have stouter abdomens than female castes, therefore, their abdomen width is expected to be larger (WOYKE, 1978). In the last decade some authors studied morphometric at the queen's emergence in an Africanized population. Costa (2005), reported 9.9 ± 0.58 mm for abdomen length and 4.6 ± 0.04 mm for abdomen width. Costa-Maia (2009), found 10.61 ± 0.97 mm for length and 4.96 ± 0.44 mm for width. Halak (2012), stated 11.65 ± 0.9 mm for abdomen length and 5.21 ± 0.41 mm for abdomen width, and Martins (2014), pinpointed 10.60 ± 0.87 mm for length and 4.89 ± 0.38 mm for the abdomen width. Rodrigues (2016) reported at maturity, 15.37 ± 0.91 mm for total length, 7.69 ± 0.68 mm for abdomen length, 5.50 ± 0.48 mm for abdomen width, 12.36 ± 0.96 mm for wing length, and 3.86 ± 0.61 mm for wing width.

Nonetheless, in a breeding program it is important to consider several traits simultaneously. The study of genetic correlations represents a great role in breeding programs since the phenotypic correlations *per se* do not properly represent the magnitude of genetic and environmental components. On the other hand, the genetic correlations allow verifying the probability of two different traits being affected by the same genes (PEREIRA, 2012). Understanding the

magnitude and direction of genetic correlations can assist in selection decisions. Consequently, traits that are easier to measure or require fewer resources but that show favourable genetic correlations with economic important but complex to measure traits can be used as indicator traits in selection criteria.

2.3.3 Mucus glands and seminal vesicle

The study of the mucus gland is relevant because the mating sign that each drone leaves when mating with a queen essentially consists of mucus gland proteins (COLONELLO & HARTFELDER, 2005). Martins (2014) found similar estimates for Africanized honeybee queens with a genetic correlation between weight at emergence and ovarian weight of 0.49. These estimated values are moderate but represent a starting point to evaluate the additive gene action and interaction between the weight and reproductive traits for queens and drones.

In order to correlate the morphometric characteristics of *Apis mellifera jemenitica* and *Apis mellifera carnica* at emergence and sperm number at 14 days, El-Kazafy and Abdulaziz (2013) found positive phenotypic correlations between body weight and: testicle size (0.99), seminal vesicle size (0.99), and mucus gland size (0.96). The same authors pointed out that, the size of the seminal vesicle and mucus gland may be related to the testicle size, showing a positive phenotypic correlation of 0.99 and 0.97, respectively. When correlated, the number of spermatozoa to the size of the testis, mucus gland and seminal vesicle, the data obtained were 0.99, 0.99 and 0.97 respectively. Indicating that body weight may serve as a selection parameter for obtaining larger reproductive organs and consequently greater production of spermatozoa.

In addition to the size of the reproductive organs, the sperm volume of Africanized *Apis mellifera* can be influenced by environmental factors, reaching its peak in the spring, decreasing gradually as summer passes and autumn arrives. However, the bumblebees produced in the fall period have a higher concentration of spermatozoa in the semen than the drones of other seasons (RHODES, 2002).

Estimation of genetic parameters of morphometric traits and reproductive organs of drone bee is fundamental in any apicultural production system since the quality of drone is directly related to the reproductive performance.

3 Material and methods

3.1 Study area

The study was carried out within the *Unidade de Ensino e Pesquisa de Apicultura* (UNEPE) of the federal university of technology - Panará, Dois Vizinhos, Brazil (latitude: - 25.70°; longitude: - 53.10°; altitude: 546 m).

3.2 Rearing of drones

Six colonies chosen randomly with identified queens were used to rear drones of an Africanized bee's population. This population had been selected by genetic values of the queen's weight, two years before the start of this study. During the drone-rearing period no genetic selection took place, therefore this population was considered as under relaxed selection (LAHTI et al., 2009).

All drones were reared in Langstroth hives according to the methodologies used by Williams and Free (1975) and Boes (2010). Each hive with 10 frames (6 brood frames covered with bees and 4 food frames), was weekly fed with protein supplement described by Sereia (2009) and sugar syrup (water and sugar, 1:2 v/w). Before introducing a frame with drawn drone wax, all the drones in each colony were killed as suggested by Boes (2010), since the drone laying activity dependent on the number of drones already present inside the hive. We introduced one frame per colony, between two frames with capped brood and checked for bee numbers and sanitary state as to guarantee a uniform pattern. Every three days all the drone frames were inspected to confirm the presence of eggs and larvae and predict an emergence time. Twenty-three days after detecting drone eggs, the frames were taken to a controlled humidity (60%) and temperature (34ºC) incubator and a mesh was used to keep the frames in (Picture 1). All frames were monitored until all the drones had emerged.

Source: Rodrigues, 2016

Picture 1: Frame with drone combs at the level of apiary and frame with drone combs inside the incubator in a fabric mesh

3.3 Data collection

Experimental dataset was obtained from October 2013 to April 2014, spring-summer season. The dataset was comprised of a total of 329 individuals drone records at maturity (Figure 3). Data collected were about: weight at maturity, total length, length of abdomen, width of abdomen, length of wing, width of wing, weight of seminal vesicle, area of mucus gland, area of seminal vesicle, volume of mucus gland and volume of seminal vesicle.

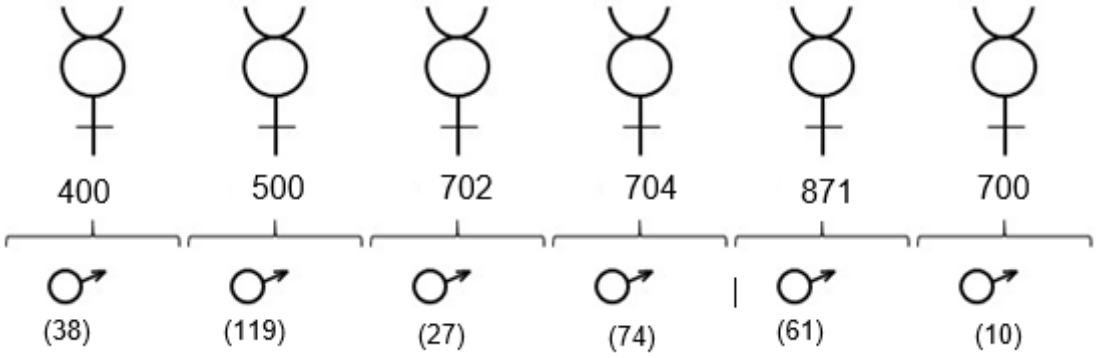
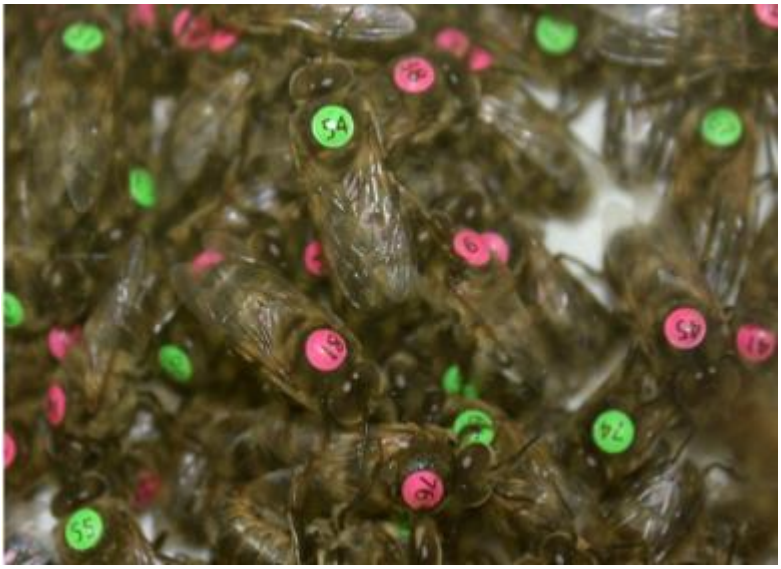


Figure 3: Matrices and their representations in the dataset

3.3.1 Morphometric data

After emergence, all the drones were anesthetized with CO_2 in an adapted chamber and then each identified with an individual numbered and colored opalite marker (Picture 2). Biometric data were recorded and drones were introduced into queen less colonies with five frames with capped and open brood, high population and with no drones inside (FREE, 1957; WILLIAMS; FREE, 1975; WHARTON et al., 2007; WHARTON et al., 2008; BOES, 2010). These hives were

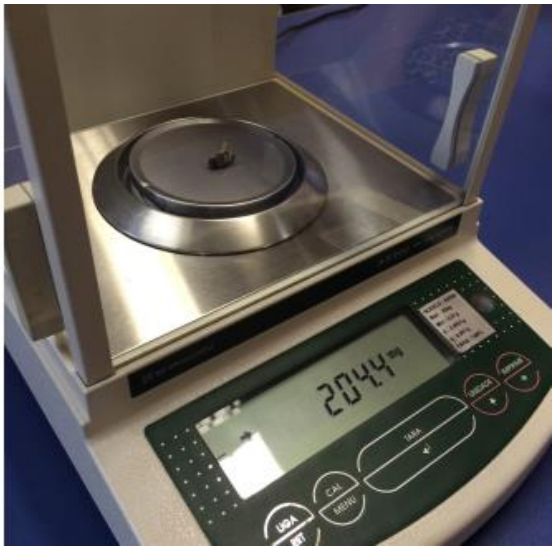
fed weekly with protein powder developed by Sereia (2009) and sugar solution (1:1, w/v).



Source: Rodrigues, 2016

Picture 2: Identified drones with an individual numbered and colored opalite marker, at emergence

On the 24th day after emergence, considered as the maturity age (RHODES, 2008) the drones were caught and taken to lab in a Styrofoam box with workers, to record their weight and body measures. All the drones were anesthetized with CO_2 in an adapted chamber. Weight was obtained with a precision scale balance (Picture 3) and body measures were measured with a caliper ruler (Picture 4).



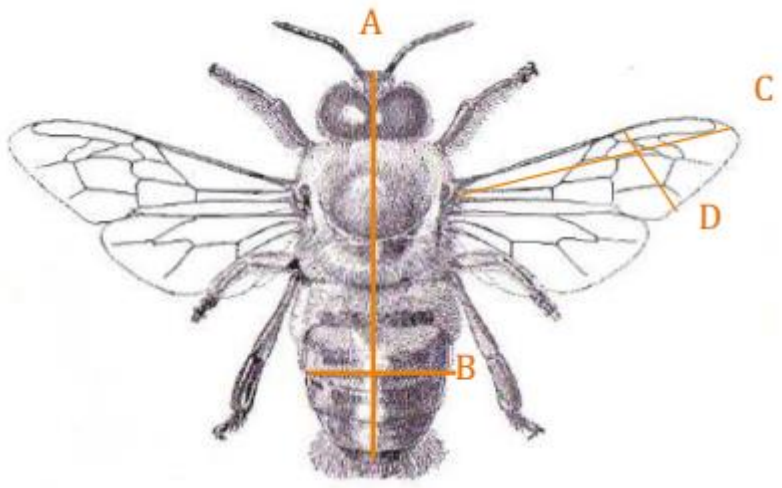
Source: Rodrigues, 2016

Picture 3: Precision scale balance (0.0001g) Shimadzu/AX 200

Source: Rodrigues, 2016

Picture 4: Caliper ruler

The weight (mg), total length (mm), abdomen length (mm), abdomen width (mm), wing length (mm), and wing width (mm) were recorded (Figure 4).

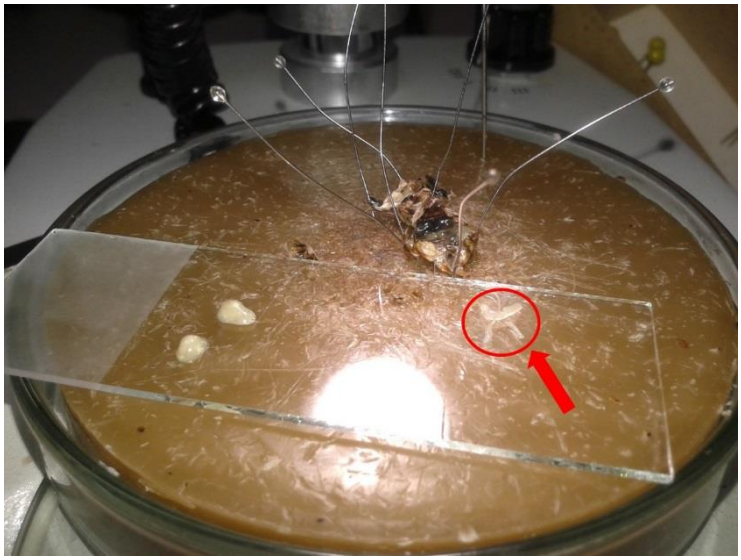


Source: Adapted from Dade (1994)

Figure 4: An outline of the different segments measured (A) Total length, (B) Abdomen width, (C) Wing length and (D) Wing width

3.3.2 Reproductive organs

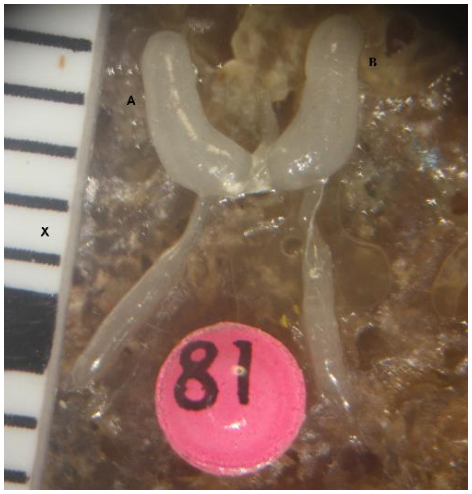
After collection of morphometric data, all the drones were anesthetized again with CO_2 in an adapted chamber drones, followed by withdrawal of their limbs (head, legs and wings), keeping the thorax and abdomen. The remaining parts were fixed with the help of entomological pins in a Petri dish containing beeswax and dissected using a forceps (Clockmaker 12 cm straight) and scissors (Ophthalmic Surgical Capsulotomy Curvature Vannas IM-283AA) under a microscope stereoscopic binocular (Quimis / Q714Z-1). For the exposure of the mucus gland and seminal vesicle, entomological pins adapted to an acrylic handle and a glass slide (Picture 5) were used.



Source: Anonymous

Picture 5: Exposure of mucus gland and seminal vesicles of drone

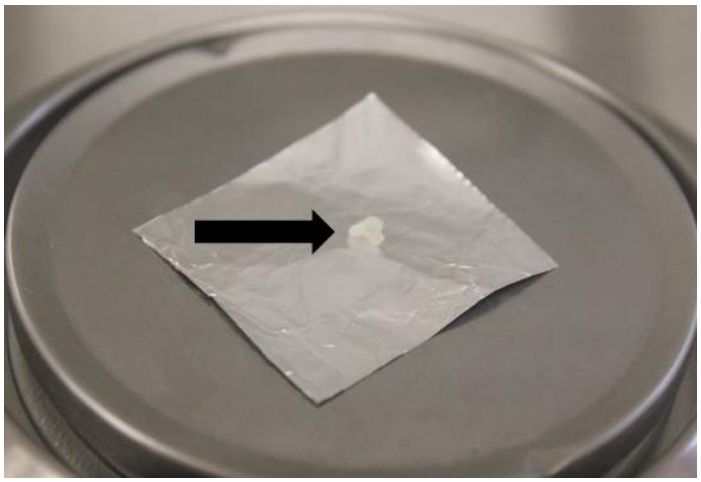
Once exposed, the mucus gland and seminal vesicle followed a standard order of positioning for photographic recording. In addition to the standard positioning, the photos were taken in the presence of a millimeter ruler with a reference mark and a numbered plaque, identifying the one to which the proper organ belongs (Picture 6).



Source: Anonymous

Picture 6: Photographic pattern including the position of the mucus gland and seminal vesicles of africanized *Apis mellifera* drone; millimeter ruler with reference point and numbered plate

After photographic recording, the weighing of the mucus gland and seminal vesicles was performed with a precision scale balance (Shimadzu / AX-200), under an aluminium foil, which was weighed at each weighing of the reproductive organs (Picture 7).



Source: Anonymous

Picture 7: Weighing of the reproductive organs of africanized *Apis mellifera* drone

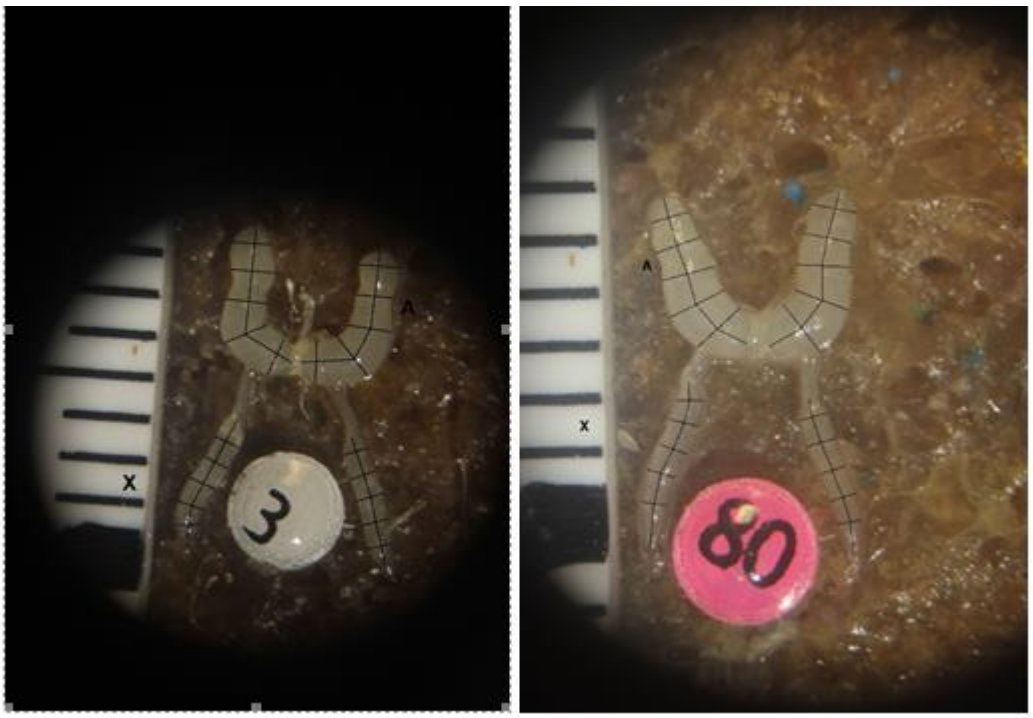
To estimate the area and the volume of the mucus gland and seminal vesicles, the UTHSCA image tool program, version 3.0 was used. The steps followed were: calibration of the ruler by the means of a reference point in the

image (standard mark on the millimeter ruler); manual contour in each of the gland, measuring side A and side B separately; recording of final values. The mucus gland and the seminal vesicles of the drones present cylindrical shape (SNODGRASS, 1956), making possible the estimation of the area and the volume by the means of the following formula:

$$V = [\pi \frac{(\frac{L_{1_{la}}+L_{2_{la}}+L_{3_{la}}+L_{4_{la}}+L_{5_{la}}}{5})^2}{2} * CT_{la}] + [\pi \frac{(\frac{L_{1_{lb}}+L_{2_{lb}}+L_{3_{lb}}+L_{4_{lb}}+L_{5_{lb}}}{5})^2}{2} * CT_{lb}]$$

Where: V= Estimated volume; $L_{1_{la}}$ to $L_{5_{la}}$= Abdomen width of side a; CT_{la}= Total length of side a; $L_{1_{lb}}$ to $L_{5_{lb}}$= Abdomen width of side b; CT_{lb}= Total length of side b

Measurements of width and length were made with the help of the UTHSCA image tool, version 3.0 (Picture 8).



Source: Anonymous

Picture 8: Measurement of the mucus gland and seminal vesicles of *Apis mellifera* Africanized drones, through the UTHSCA image tool program, version 3.0

3.4 Data analysis

Single-trait and two-trait models were used and parameters such as components of variance, heritability and genetic correlations were estimated using BLUPF90 family of program.

Two distinct fixed effects were considered: the hives where drones were kept from emergence until maturity, and the yearly time (three times throughout the year: end of spring, and in the beginning and end of summer).

Additive genetic effect and residual error were assumed as random effects and normal distribution was assumed, except for genetic (co)-variance components where inverted Gama and Wishart distribution were considered. Bayesian estimation was obtained through a Gibbs sampling method.

Genetic parameters were estimated considering the father as unknown based on BLUP - Animal Model approach as follows:

$$y = X\beta + Za + e$$

where *y* is the vector of records; *X* is the incidence matrix relating the observations to the corresponding environment, contained in the vector *β*; *β* is the vector of fixed period/hive effects; *Z* is an incidence matrix of additive genetic effects; *a* is a vector of additive genetic effects; and *e* is the vector of random errors associated to each observation.

Normal multivariate joint distribution was assumed for the vectors *y*, *a* and *e*:

$$\begin{bmatrix} y \\ a \\ e \end{bmatrix} \sim NMV\left\{\begin{bmatrix} X\beta \\ 0 \\ 0 \end{bmatrix};\begin{bmatrix} ZGZ'+R & ZG & R \\ GZ' & G & 0 \\ R & 0 & R \end{bmatrix}\right\}$$

Single-trait analysis

G represents the genetic (co)-variance matrix as $A\sigma_a^2$. *A* represents the numerator relationship matrix which indicates the additive genetic relationship between individuals which is symmetric and its diagonal element for animal. *i* is equal to *1* + F_i where F_i is the inbreeding coefficient of animal *i* (WRIGHT, 1922), and σ_a^2 is the additive genetic variance; *R* is the residual variance matrix given by $I\sigma_e^2$, and *I* represent the identity matrix with order equal to the number of drones,

and σ_e^2 is the residual variance for each trait.

Two-trait analysis

The *G* matrix is given by $G_0 \otimes A$, *A* being the relationship matrix, and G_0 is the matrix of genetic (co)-variance as follows:

$$G_0 = \begin{bmatrix} \sigma_{a_1}^2 & \sigma_{a_1 a_2} \\ \sigma_{a_2 a_1} & \sigma_{a_2}^2 \end{bmatrix}$$

The R matrix is given by $R_0 \otimes I$, where *I* represents the identity matrix with order equal to the number of drones and R_0 is the residual variance matrix for each trait given as follows:

$$R_0 = \begin{bmatrix} \sigma_{e_1}^2 & \sigma_{e_1 e_2} \\ \sigma_{e_2 e_1} & \sigma_{e_2}^2 \end{bmatrix}$$

Sampling

Considering the univariate analysis (single-trait) and the multivariate analysis (multi-trait), probabilities from scalar-*Gibbs* with a chain of length 1.000.000 were estimated including a burn-in period of 100.000 rounds, and a thinning interval of 1 for all analyses.

Heritabilities and genetic correlations for each trait were calculated as follows:

$$h^2 = \frac{\sigma_a^2}{\sigma_y^2}$$

Where,

h^2= heritability coefficient;

σ_a^2 = additive genetic variance;

σ_y^2 = phenotypic variance.

$$r_g = \frac{\sigma_{a_1 a_2}}{\sqrt{\sigma_{a_1}^2 . \sigma_{a_2}^2}}$$

Where,

r_g= genetic correlation between trait 1 and trait 2, respectively;

$\sigma_{a_1 a_2}$ = additive genetic covariance between trait 1 and trait 2, respectively;

$\sigma_{a_1}^2$ and $\sigma_{a_2}^2$ = additive genetic variance between trait 1 and trait 2, respectively.

Convergences of Gibbs sampling-chains were performed by Heidelberger and Welch (1983) diagnostic tests, which firstly compares the Gibbs chain with a hypothetical chain of stationary distribution, then verifies whether the means of the sampled data are within a threshold of the credibility interval established. These diagnostic tests and the mode of each component were tested with CODA (Convergence Diagnosis and Output Analysis) library, implemented in the R software.

The percentage of credibility intervals, and regions of high density were constructed for all the (co)-variance components at the 90% level of credibility, meaning that there is a 90% probability that the true value of θ lies within the credible region.

4 Results and discussion

4.1 Results

Mean and standard deviation for each measured trait at maturity are presented in Table 3.

Table 3: Mean and standard deviation for each measured trait at maturity

Traits	Mean and standard deviation
Weight (W)	202.81 ± 17.84 mg
Total length (TL)	15.39 ± 0.74 mm
Abdomen length (AL)	7.69 ± 0.68 mm
Abdomen width (AW)	5.48 ± 0.29 mm
Wing length (WL)	12.40 ± 0.66 mm
Wing width (WW)	3.83 ± 0.30 mm
Mucus gland weight (MGW)	12.60 ± 1.9 mg
Seminal vesicle weight (SVW)	1.80 ± 0.59 mg
Mucus gland area (MGA)	25.45 ± 8.59 mm^2
Seminal vesicle area (SVA)	8.60 ± 2.92 mm^2
Mucus gland volume (MGV)	37.84 ± 18.12 mm^3
Seminal vesicle volume (SVV)	6.65 ± 3.31 mm^3

As revealed by Table 3, the average weight at maturity is 202.81 ± 17.84 mg and the average length and width of abdomen are 7.69 ± 0.68 and 5.48 ± 0.29 mm respectively. Weight, area and volume of seminal vesicle are respectively, 1.80 ± 0.59 mg, 8.60 ± 2.92 mm^2 and 6.65 ± 3.31 mm^3.

The summary estimates of additive genetic variance ($\boldsymbol{\sigma_a^2}$), residual ($\boldsymbol{\sigma_e^2}$), phenotypic ($\boldsymbol{\sigma_y^2}$) and heritability ($\boldsymbol{h^2}$) using analysis of single-trait model with credibility intervals and regions of high density, at the 90% level of credibility, for weight (W), total length (TL), abdomen length (AL), abdomen width (AW), wing length (WL), wing width (WW), mucus gland weight (MGW), seminal vesicle weight (SVW), mucus gland area (MGA), seminal vesicle area (SVA), mucus gland volume (MGV), seminal vesicle volume (SVV) of *Apis mellifera* Africanized drones is presented in Table 4.

Table 4: Estimates of additive genetic variance ($\boldsymbol{\sigma_a^2}$), residual ($\boldsymbol{\sigma_e^2}$), phenotypic ($\boldsymbol{\sigma_y^2}$) and heritability ($\boldsymbol{h^2}$) using analysis of single-trait model with credibility intervals and regions of high density, at the 90% level of credibility, for weight (W), total length (TL), abdomen length (AL), abdomen width (AW), wing length (WL), wing width (WW), mucus gland weight (MGW), seminal vesicle weight (SVW), mucus gland area (MGA), seminal vesicle area (SVA), mucus gland volume (MGV), seminal vesicle volume (SVV) of *Apis mellifera* Africanized drones

	Parameter			
Traits	σ_a^2	σ_e^2	σ_y^2	h^2
W[1]	164.20 (39.98 – 321.10)[+] (26.29 – 302.80)[++]	140.38 (22.48 – 241) (22.38 – 241)	304.58 (256.80 – 360.45) (253.58 – 356.02)	0.52 (0.14 – 0.93) (0.16 – 0.94)
TL[2]	0.44 (0.21 – 0.64) (0.22 – 0.66)	0.15 (0.01 – 0.34) (0.000 – 0.29)	0.59 (0.51 – 0.69) (0.50 – 0.69)	0.74 (0.38 – 0.98) (0.47 – 1)
AL[2]	0.26 (0.06 – 0.51) (0.05 – 0.49)	0.21 (0.03 – 0.38) (0.03 – 0.37)	0.48 (0.40 – 0.56) (0.39 – 0.56)	0.54 (0.14 – 0.94) (0.16 – 0.96)
AW[2]	0.06 (0.02 – 0.10) (0.01 – 0.09)	0.03 (0.00 – 0.06) (0.000 – 0.06)	0.09 (0.08 – 0.11) (0.07 – 0.10)	0.61 (0.22 – 0.96) (0.29 – 1)
WL[2]	767.84 (30.11 – 2745) (0.25 – 2008)	2494.16 (993 – 3273) (1385 – 3431)	3262.01 (2785 – 3882) (2714 – 3780)	0.22 (0.01 – 0.73) (0.000 – 0.56)
WW[2]	0.05 (0.02 – 0.09) (0.02 – 0.09)	0.03 (0.003 – 0.06) (0.000 – 0.05)	0.08 (0.07 – 0.10) (0.07 – 0.09)	0.63 (0.22 – 0.96) (0.31 – 1)
MGW[1]	2.13 (0.34 – 4.39) (0.12 – 4.06)	1.99 (0.29 – 3.52) (0.20 – 3.42)	4.12 (3.31 – 5.09) (3.27 – 4.99)	0.50 (0.09 – 0.94) (0.11 – 0.95)
SVW[1]	0.22 (0.02 – 0.44) (0.000 – 0.40)	0.17 (0.02 – 0.34) (0.001 – 0.32)	0.40 (0.32 – 0.49) (0.31 – 0.48)	0.54 (0.07 – 0.96) (0.13 – 1)
MGA[3]	21.91 (0.88 – 66.55) (0.001 – 55.06)	43.93 (9.95 – 65.84) (14.04 – 68.67)	65.84 (52.03 – 83.15) (50.27 – 80.75)	0.31 (0.01 – 0.87) (0.000 – 0.75)
SVA[3]	2.92 (0.10 – 8.44) (0.000 – 7.17)	5.25 (1.05 – 8.16) (1.33 – 8.41)	8.17 (6.31 – 10.51) (6.11 – 10.21)	0.34 (0.01 – 0.88) (0.000 – 0.78)
MGV[4]	129.66 (8.62 – 326.60) (0.09 – 284.50)	181.52 (32.05– 297.40) (32.25 – 297.50)	311.18 (242.27 – 396.60) (235.20 – 386.50)	0.40 (0.03 – 0.91) (0.000 – 0.82)
SVV[4]	6.86 (1.28 – 12.68) (0.81 – 12.06)	4.43 (0.45– 9.29) (0.21 – 8.30)	11.29 (8.54 – 14.68) (8.22 – 14.21)	0.59 (0.13 – 0.96) (0.21– 1)

[1] milligram; [2] millimeter; [3] square millimeter; [4] cubic millimeter; [+] credibility interval at the 90% level of credibility; [++] region of high density at the 90% level of credibility

It stems from Table 4 that, heritability of traits varies from 0.22 to 0.74. Heritabilities of weight, area, and volume of mucus gland are respectively, 0.50,

0.31 and 0.40. Heritabilities were considered low when values fell within the range of 0.10-0.29, moderate when within the range of 0.30-0.49 and high when they equaled or exceeded 0.50.

Table 5 shows, the estimates of genetic correlations $(\mathbf{r}_{\mathbf{g}_{1,2}})$ for weight (W), total length (TL), abdomen length (AL), abdomen width (AW), wing length (WL), wing width (WW), and mucus gland weight (MGW), seminal vesicle weight (SVW), mucus gland area (MGA), seminal vesicle area (SVA), mucus gland volume (MGV), seminal vesicle volume (SVV) of *Apis mellifera* Africanized drones.

Table 5: Estimates of genetic correlations $(\mathbf{r}_{\mathbf{g}_{1,2}})$ for weight (W), total length (TL), abdomen length (AL), abdomen width (AW), wing length (WL), wing width (WW), and mucus gland weight (MGW), seminal vesicle weight (SVW), mucus gland area (MGA), seminal vesicle area (SVA), mucus gland volume (MGV), seminal vesicle volume (SVV) of *Apis mellifera* Africanized drones

$r_{g_{1,2}}$	W[1]	TL[2]	AL[2]	AW[2]	WL[2]	WW[2]
MGW[1]	-0.07 (-0.87 – 0.76)[+] (-1 – 0.63)[++]	-0.22 (-0.93 – 0.50) (-1 – 0.37)	-0.43 (-0.99 – 0.47) (1 – 0.28)	-0.20 (-0.94 – 0.61) (-1 – 0.46)	0.18 (-0.95 – 0.99) (-0.85 – 1)	-0.46 (-1 – 0.46) (-1 – 0.28)
SVW[1]	-0.39 (-0.99 – 0.51) (-1 – 0.33)	-0.72 (-1 – -0.07) (-1 – -0.25)	-0.72 (-0.99 – 0.06) (-1 – -0.19)	-0.54 (-1 – 0.31) (-1 – 0.11)	0.03 (-0.97 – 0.99) (0.94 – 1)	-0.78 (-1 – -0.16) (-1 – -0.37)
MGA[3]	-0.31 (-1 – 0.84) (-1 – 0.65)	-0.11 (-0.97 – 0.88) (-1 – 0.71)	-0.02 (-0.99 – 0.98) (-1 – 0.92)	-0.16 (0.99 – 0.90) (-1 – 0.74)	-0.99 (-1 – -0.98) (-1 – -0.99)	0.07 (-0.97 – 0.99) (-0.86 – 1)
SVA[3]	-0.05 (-0.98 – 0.97) (-1 – 0.88)	-0.21 (-0.96 – 0.86) (-1 – 0.66)	0.01 (-0.98 – 0.99) (-0.92 – 1)	-0.001 (-0.96 – 0.98) (-0.87 – 1)	-0.99 (-1 – -0.99) (-1 – -0.99)	-0.12 (-0.99 – 0.98) (-1 – 0.93)
MGV[4]	-0.29 (-0.99 – 0.70) (-1 – -0.52)	-0.36 (-0.99 – 0.53) (-1 – 0.33)	-0.31 (-0.99 – 0.83) (-1 – 0.61)	-0.11 (-0.99 – 0.88) (-1 – 0.72)	-0.99 (-1 – -0.99) (-1 – -0.99)	-0.24 (-0.99 – 0.79) (-1 – 0.61)
SVV[4]	-0.05 (-0.88 – 0.84) (-1 – 0.68)	-0.53 (-0.99 – 0.11) (-1 – -0.04)	-0.42 (-0.99 – 0.47) (-1 – 0.26)	-0.16 (-0.87 – 0.63) (-1 – 0.46)	-0.99 (-1 – -0.99) (-1 – -0.99)	-0.61 (-1 – 0.15) (-1 – -0.06)

[1] milligram; [2] millimeter; [3] square millimeter; [4] cubic millimeter; [+] credibility interval at the 90% level of credibility; [++] region of high density at the 90 level of credibility

It follows from Table 5 that, genetic correlations between morphometric traits and reproductive organs varies from -0.99 to 0.18. These values are respectively, the genetic correlations between, wing length and seminal vesicle volume, and wing length and mucus gland weight. To certain limits, the size of the reproductive organs seems not to depend on the size of the body and morphometric traits.

4.2 Discussion

In breeding programs where instrumental insemination is used, it is very important to set criteria for selection of drones that belong to known genetic sources. One method to achieve that is to estimate genetic parameters of morphometric traits and reproductive organs of drones at mature stage. From our results, it appears that:

In terms of weight at maturity, our result collaborates with the findings of Gencer and Firatli (2005), and Rodrigues (2016). This result is superior by 5.87% to the weight of Yemeni subspecies at emergence and inferior to 12.03% to the weight of Carniolan subspecies at emergence (EL-KAZAFY and ABDULAZIZ, 2013). These differences confirm the fact that, the weight of bees varies with endogenous factors (species, subspecies, sex, age…) and exogenous factors (period of the year, availability of food…). This weight decrease might be explained through the testes involution process, after the first week of adult life, during which the sperm migrates from the testes to the seminal vesicles where they undergo the final stages of maturation. The decrease in weight may be regarded as an indicator of reaching maturation.

Considering the wing length and width, our result is comparable to that which was reported in German by Schlüns et al. (2003) and in Saudi Arabia by El-Kazafy and Abdulaziz (2013) for Yemeni subspecies. On the other hand, our result is inferior by 11.49 and 8.15% respectively for wing length and width, when compared to Carniolan subspecies. This difference could be explained by the genetic composition between subspecies and the environmental factors to which the bee populations have been subjected.

In terms of total length, width and length of abdomen, our results are inferiors to 1.1, 2.01 and 3.77% respectively when compared to results reported by Rodrigues (2016) for a newly emerged drone. This may be due to the fact that,

the abdomen of a newly emerged drone is large because of testes, which lie in the expanded abdomen, are still filled with spermatozoa (MAZEED and MOHANNY, 2010). As the spermatozoa move to the seminal vesicles, the testes shrink and finally become amorphous tissue. This is accompanied by reduction in size of the abdomen. This result reminds us of their importance during evaluation of drone body sizes and therefore evaluation of their internal contents. It is also important from the selection point of view. Regarding the difference of the abdomen width, it could also be due to the availability of food. Abdomen length and width are relevant because that is where the reproductive organs are located and hence it can be used for evaluating the drone's growth and body development.

According to Rinderer et al. (1985), the weights of mucus gland and seminal vesicle for Africanized drones are respectively 14.4 and 1.5 mg. The average weight of mucus gland recorded in Venezuela by Rinderer et al. (1985) for European drones is 12.70% slightly larger than our result. This may be explained by the fact that, weight of seminal vesicle increase with age until maturity and then start to decrease, certainly due to absorption. The precise role of mucus in natural mating is unclear. It does become involved in the formation of a "mating sign" and may be involved in competition among drones for greater representation of their spermatozoa in the final contents of queen's spermatheca (RINDERER et al., 1985).

The volume of mucus gland and seminal vesicle for *Apis mellifera* Africanized drones bees at maturity were about 91.20 and 87.07% higher than those of Yemeni subspecies respectively. They were also 88.16 and 79.85% higher than those of Carniolan subspecies ones, respectively as recorded by El-Kazafy and Abdulaziz (2013) in Saudi Arabia. This may be explained by variation in sizes of body, testis and seminal vesicle, and exogenous factors. To certain limits, the size of the reproductive organs seems not to depend on the size of the body (MAZEED and MOHANNY, 2010). Thus, the size of seminal vesicle in the matured drones does not help to predict the real quantity and quality of spermatozoa.

With regards to the heritability, it appears that, it varies from 0.22 to 0.74. Heritability for *Apis mellifera* Africanized queen at emergence, reported by Martins (2014), vary from 0.46 to 0.80. This difference possibly resulted from

haploid nature of drones. This study revealed that, traits studied presented genetic variability needed for preservation and genetic improvement by conventional tools such as selection and breeding.

In this study, genetic correlations between morphometric traits and reproductive organs varies from -0.99 to 0.18. Schlüns et al. (2003) found a significant positive phenotypic correlation (0.49) between sperm number and wing length within the small drones. El-Kazafy and Abdulaziz (2013), reported significant positive phenotypic correlations between mean volume of testis, seminal vesicle and mucus gland in one hand and mean weight of drone (r =0.99, 0.99, 0.96; P< 0.01), length of fore wing (r = 0.99, 0.98, 0.97; P< 0. 01), width of forewing (r = 0.99, 0.98, 0.98; P< 0.01), length of hind wing (r = 0.85, 0.80, 0.84; P< 0.05), and number of hamuli (r = 0.94, 0.91, 0.92; P< 0.01) on the other hand, respectively. This result reminds us of the necessity to consider genetic correlations instead of phenotypic correlations in a breeding program, because environmental component influence significantly phenotypic correlations. Indeed, such genetic correlations are necessary for the indication of potential relationships among characters. It would be helpful for the selection and breeding programs.

5 Conclusion

Base on this study on genetic aspects of morphometric traits and reproductive organs of Africanized honey bee drones, *Apis mellifera L.* (Hymenoptera: Apidae), it could be concluded that:

Morphometric traits and reproductive organs when evaluated separately can be used as selection criteria.

Selection for wing length associated with mucus gland weight should result in a low genetic progress.

To certain limits, the size of the reproductive organs at maturity seems not to depend much on the size of the body and morphometric traits considered in this study.

6 REFERENCES

BERG, S. KOENIGER, N. KOENIGER, G AND FUCHS, S. body size and reproductive success of drones (*Apis mellifera* L.). **Apidologie** 28, 449-460, 1997.

BIENEFELD, K. EHRHARDT, K. REINHARDT, F. Genetic evaluation in the honey bee considering queen and worker effects - a BLUP - animal model approach. **Apidologie**, v. 38, p. 77-85, 2007.

BIENEFELD, K. PIRCHNER, F. Heritabilities for several colony traits in the honeybee (*Apis mellifera carnica*). **Apidologie**, v. 21(3), p. 175-183, 1990.

BOES, K. Honeybee colony drone production and maintenance in accordance with environmental factors: an interplay of queen and worker decisions. **Insectes Sociaux**, V. 57(1), p. 1–9, 2010.

BRASCAMP, E. BIJMA, P. Methods to estimate breeding values in honey bees. **Genetics, Selection, Evolution**, v. 46(1), p. 53, 2014.

CALDERONE, N. Insect pollinated crops, Insect pollinators and US agriculture: Trend analysis of aggregate data for the period 1992-2009*.* **PLos ONE**, 7, e 37235, 2012.

COLONELLO, N. HARTFELDER, K. She's my girl - male accessory gland products and their function in the reproductive biology of social bees. **Apidologie,** v. 36, p. 231–244, 2005.

COSTA, F.M. **Estimativas de parâmetros genéticos e fenotípicos para peso e medidas morfométricas em rainhas *Apis mellifera* africanizadas**. 2005. 39 f. Dissertação (Mestrado em Zootecnia) - Programa de Pós Graduação em Zootecnia, Universidade Estadual de Maringá, Maringá, 2005.

COSTA-MAIA, F. M et al. Estimates of covariance components for hygienic behavior in Africanized honeybees (*Apis mellifera*). **Revista Brasileira de Zootecnia**, v. 40, n. 9, p.1909-1916, 2011.

COSTA-MAIA, F. **Aspectos genéticos da produção de mel e comportamento higiênico em abelhas *Apis mellifera* africanizadas**. Tese (Doutorado em Zootecnia) – Programa de Pós Graduação em Zootecnia, Universidade Estadual de Maringá, Maringá. 77p, 2009.

CULLINEY, T. W. Origin and evolutionary of the honeybees *Apis*. **Bee World**, 64: 29-37, 1983.

DADE, H. Anatomy and dissection of the honeybee. **The Alden Press, Oxford**, UK, 158p, 1994.

DUAY, P. JONG, D. ENGELS, W. Decreased flight performance and sperm production in drones of the honeybee (Apis mellifera) slightly infested by Varroa destructor mites during pupal development. **Genetic Molecular Research,** 1 (3), 227-232, 2002.

El-Kazafy A.T. and Abdulaziz S.A. Morphometric and Reproductive Organs Characters of *Apis mellifera jemenitica* Drones in Comparison to *Apis mellifera carnica*. **International Journal of Scientific & Engineering Research**, Volume 4, Issue 10, 2013.

EVERT, B. WILLAM, A. BOIGENZAHN, C. BIJMA, P. VEERKAMP, R. Heritabilities and genetic correlations for honey yield, gentleness, calmness and swarming behaviour in Austrian honey bees. **Apidologie,** 47:739–748, 2016.

FOOD AND AGRICULTURE ORGANIZATION (FAO). Country fact sheet on food and agriculture policy trends, 2014.

FREE, J. The food of adult drone honeybees (*Apis mellifera*). **The British Journal of Animal Behaviour**, v. 5, p. 7–11, 1957.

GENCER H.V, FIRATLI C. Reproductive and morphological comparisons of drones reared in queen right and laying worker colonies. **Journal of Apicultural Research**, 44: 163–167. 2005.

GUPTA, P. REINSCH, N. SPÖTTER, A. CONRAD, T. BIENEFELD, K. Accuracy of the unified approach in maternally influenced traits illustrated by a simulation study in the honeybee (Apis mellifera). **BMC Genetics**, V. 14 (36), P. 36, 2013.

HALAK, A.L. **Parâmetros e correlações genéticas e fenotípicas para peso e medidas morfométricas em rainhas *Apis mellifera* africanizadas**. 2012. 46 f. Dissertação (Mestrado em Zootecnia) - Programa de Pós Graduação em Zootecnia, Universidade Estadual de Maringá, Maringá, 2012.

HEIDELBERGER, P. WELCH, P.D. Simulation run length control in the presence of an initial transient. **Operations Research**, v. 31, n. 6, p. 1109-1144, 1983.

KERR, W.E. Método de seleção para melhoramento genético em abelhas. **Magistra**, v.18, n. 4, p. 209-212, 2006.

LAHTI, D. Relaxed selection in the wild. **Trends in Ecology and Evolution**, v. 24(9), p. 487–496, 2009.

LAIDLAW, H.R AND PAGE, R, E. Queen rearing and bee breeding. **Wicwas press, Cheshire,** 1998.

MARTINS, J. **Aspectos genéticos de caraterísticas morfométricas e reprodutivas de rainhas Apis mellifera L. (Hymenoptera: Apidae) africanizadas.** Dissertação (Mestrado em Zootecnia) Programa de Pós Graduação em Zootecnia - Universidade Tecnológica Federal do Paraná, Dois Vizinhos, 89p, 2014.

MAZEED, A. M. AND MOHANNY, K. M. Some reproductive characteristics of honeybee drones in relation to their ages. **Entom. Res**.40 (5), 240-250, 2010.

MICHENER, C. D. The social behavior of the bees: a comparative study. **Harvard Univ. Press**. Cambridge, Mass, 56p, 1974.

MICHENER, C. The bees of the world (second). Maryland: **the Johns Hopkins University press**, 953pp, 2007.

MOSTAJERAN, M et al. Heritability estimates of morphological characters and honey yield of honeybee colonies in Isfahan. **Journal of Science & Technology of Agriculture and Natural Resources**, v. 4, p. 119-126, 2000.

OTTO, M. KENNETH W. TUCKER. Instrumental insemination of queen bees. **Entomology Research Division, Agricultural Research Service**, 1970.

PADILHA, A. et al. Genetic parameters for five traits in Africanized honeybees using Bayesian inference. **Genetics and Molecular Biology**, v. 36, n. 2, p. 207-213, 2013.

PEREIRA, J. Melhoramento genético aplicado à produção animal. **Belo Horizonte: FEPMVZ**, 758p, 2012.

RHODES, J. Drone honeybees rearing and maintenance. NSW Agriculture DAI/112 ISSN 1034-6848 Ed. **William E. Smith,** 2002.

RHODES, J. Semen production in drone honeybees. **Rural Industries Research and Development Corporation**, v. 130(08), 95p, 2008.

RINDERER, T.E. COLLINS, A.M. PESANTE, D. A Comparison of Africanized and European drones: weights, mucus gland and seminal vesicle weights, and counts of spermatozoa. **Apidologie,** 16 (4), 407-412, 1985.

Rodrigues, M. **Genetic parameters for morphometric traits of africanized honeybee drones.** Dissertação (Mestrado em Zootecnia) - Programa de Pós-Graduação em Zootecnia - Universidade Tecnológica Federal do Paraná, Dois Vizinhos, 88p, 2016.

ROFF, C. The honey bee colony. **Queensland Agricultural Journal**, 1977.

SCHLÜNS, H. SCHLÜNS, E.A. VAN PRAAGH, J. AND MORITZ, R.F.A. Sperm numbers in drone honeybees (Apis mellifera) depend on body size. **Apidologie**, 34, 577-584, 2003.

SEREIA, M. **Suplementos proteicos para abelhas africanizadas submetidas à produção de geleia real**. Dissertação (Mestrado em Zootecnia) - Universidade Estadual de Maringá, Maringá, 92p, 2009.

SNODGRASS, R. Anatomy of the honeybee. New York. **Comstock Publish**. Ithaca, N.Y., 334p, 1956.

TCHOUMBOUÉ, J. PINTA, J.Y. DONGOCK, N.D. ZANGO, P. TEKOUNEGNING. Elevage intensif d'abeilles, **Université de Dschang**. 89p, 2010.

WHARTON, K et al. The honeybee queen influences the regulation of colony drone production. **Behavioral Ecology**, v. 18(6) p. 1092–1099, 2007.

WHARTON, K.; DYER, F.; GETTY T. Male elimination in the honeybee. **Behavioral Ecology**, v. 18(6) p. 1075–1079, 2008.

WILLIAMS, I; FREE, J. Factors determining the rearing and rejection of drones by the honey bee colony. **Animal Behavior**, v. 23, p. 650–675, 1975.

WINSTON M. The biology of the honey bee. **Harvard University Press. Cambridge,** MA, 1987.

WOYKE, J. Comparative biometrical investigation on diploid drones of the honeybee III. The abdomen and weight. **Journal of Apicultural Research**, v. 17(4), p. 206–217, 1978.

WRIGHT, S. Coefficients of inbreeding and relationship. **The American Naturalist**, v. 56, p. 330–338, 1992.