Iranian Journal of Animal Biosystematics (IJAB)
Vol.17, No.2, 157-167, 2021
DOI: 10.22067/ijab.2022.69007.1004

IJAB
ISSN: 1735-434X (print); 2423-4222 (online)

RESEARCH ARTICLE **Open access**

Male and female genital allometry in *Habrobracon hebetor* (Hymenoptera: Braconidae)

Mohammad Mahdi Rabieh[1,*] and Farzaneh Afkhami[2]

[1]*Department of Plant Protection, Faculty of Agriculture, University of Birjand, Birjand, Iran.*
[2]*Department of Plant Protection, Faculty of Agriculture, Institute of Higher Education Jahad Daneshgahi, Kashmar, Iran.*

(Received: *22 February 2021*; Accepted: 10 *December 2021*)

Abstract
This is particularly apparent within the insects that show high variations in genitalic form between closely related species. Static allometry is one of the effective approaches for quantification of such variation. Despite the crucial roles of the parasitic Hymenoptera in the ecosystems, little is known about the sexual selection in this order, compared with other insect orders. We examined the allometry of different morphological traits in 35 males and 35 females of *Habrobracon hebetor* (Say, 1836) (Hymenoptera: Braconidae) from a laboratory colony reared in Mashhad city, for the first time. The aim was the investigation of allometric relationships of different body traits as a way for quantification of the natural selection impacts on the different body parts. 12 genitalic and non-genitalic body parts of *H. hebetor* males and females were photographed and measured. Principal Component Analysis (PCA) was used to explore the variance of the traits and two regression analysis methods to obtain the allometric slopes. All the non-genitalic traits in male and female wasps showed isometry except pterostigma width in male wasps which showed positive allometry. In male genitalia, two traits showed strong negative allometry and one trait showed isometry. Our findings showed that in this species males with an average size of genitalia were more successful in generating viable offspring than males with relatively smaller or larger genitalia sizes and this is irrespective of the overall body size. Our results showed for the first time that such stabilizing sexual selection might operate on genital size in the braconid wasps.

Key words: Insect's evolution, Sexual selection, *Habrobracon hebetor*, Intraspecific variety, Morphology.

INTRODUCTION

Extraordinary variation of genitalia structures in animals and its function in the natural selection and speciation is highly considered and discussed, lately. Sexual selection is discussed as the primary force driving such enormous variation (Leonard & Córdoba-Aguilar, 2010; Cao et al. 2019). Static allometry, which is defined as the size relationship of a specific body part with the whole body in a population of a particular species, is one of the effective approaches for quantification of the natural selection impacts on the different body parts and their performance in an organism (Hosken, & Stockley, 2004; Ohno et al. 2003). This relationship is revealed by the slope attained in log-log regression of the size of a given body trait on an indicator of total body size (Eberhard, 1985). According to the selection hypotheses, relative size of different body parts is

Corresponding Author: *mmrabie@birjand.ac.ir*

examined by using an index for the overall body size. Several studies on genital allometry used a single somatic trait as the indicator of body size (e.g. pronotum length, elytra length and head width), although the use of a different body-size indicator may give rise to different results (Green, 1999). A slope over 1.0 (positive allometry) indicates advantage of relatively larger traits in the larger individuals of the population. Slope around 1.0 (isometry) shows relatively constant size of the trait across the population. Allometric slopes lower than 1.0 (negative allometry) are the indicator of relatively smaller trait expression in larger individuals (Eberhard, 2009).

Presumably, natural selection can adjust allometric slopes and consequently, affects the traits performance (Eberhard et al. 1998). Stabilizing selection is one of the main ways that selection can affects animals. In this type of selection, individuals with intermediate trait size will be favored in the population instead of exaggerated trait size which will be favored in the disruptive selection phenomena (Hosken & Stockley, 2004).

According to the selection hypotheses, when trait size increases faster than overall body size, there is a positive allometric relationship, as seen in insect's weapons. An isometric relationship exists when selection pressure is equal on the trait and overall body size, like most somatic traits in insects (Eberhard et al. 1998). If the selection pressure favors intermediate values of the trait regardless of body size, it causes negative allometry (Hosken & Stockley, 2004). Apparently, this relationship is common in male genital structures of insects and spiders (Eberhard et al. 1998; Eberhard, 2009). Conversely, Bolanos et al. (2014) found an exception to the previous findings. They interpreted isometric relationship of some genital traits in the damselflies as effect of sexual conflict situation that could change scaling pattern of genitalia in these insects. Also, some similar exceptions to the general pattern proposed by Eberhard et al. (1998) were previously found (Johnson, 1995; Cayetano et al. 2011). These findings showed that genital traits may evolve under different kinds of scaling patterns in different species. Moreover, different selective pressures which genitalia may confront during different sexual selection mechanisms need further investigations.

Eberhard et al. (1998) proposed the one-size-fits-all hypothesis based on their study of allometric relationships of genital and non-genital morphological traits in many species of arthropods including insects. They indicated that all morphological traits in the male genitalia have allometric slopes significantly lower than 1.0 and lower than the slopes for non-genitalic body traits. Their finding means that sexual selection within the arthropod populations acts through stabilizing selection and males with the average genital sizes are more favored for gene transfer to the next generations. According to this hypothesis, female morphology is relatively invariable so that the consistency of male genital morphology is an adaptation to this situation. Subsequently, several studies on different orders of insects and spiders have confirmed their findings for male genital size (e.g., Schmitz et al. 2000; Iwahashi, 2001; Tatsuta et al. 2001; Eberhard, 2002; Tschinkel et al. 2003; Hosken et al. 2005; Mutanen & Kaitala, 2006; Rabieh et al. 2015; Cao & Hayashi, 2019).

Eberhard et al. (1998) found that genitalia traits of the studied females have lower allometric slopes than the median allometric slope for non-genitalic traits but this difference was not significant. According to these results, they proposed that female genitalia may show the same patterns of variation as male genitalia. But they reported less variability for female genitalia than those of non-genitalic traits (Eberhard et al. 1998; Eberhard, 2009). Afterward, a few studies (e.g., Funke & Huber, 2005; Rabieh et al. 2015; Cao & Hayashi, 2019) confirmed their findings. Although, Nava-Bolaños et al. (2012) found that genital traits did not show negative allometry in two Odonata species, in both sexes.

Despite the crucial roles of the parasitic Hymenoptera in the ecosystems and the high diversity of mating systems, little is known about sexual selection and sexual conflict in this order, compared with other insect orders. Although, the mating systems of this diverse taxonomic group are studied in-sight of how population structure influences sex allocation strategies and how to encourage their attack and control of pest populations as one of the main groups of biological control agents in the applied research (West, 2009; Boulton et al. 2015).

Allometric relationships of different body traits including genitalic parts are poorly studied in the Hymenoptera species and most allometry studies are focused on allometric relationships of non-genitalic traits, especially in the case of sexual dimorphisms investigations (e.g., Perrard et al. 2012; Benítez et al. 2013; Polilov & Makarova, 2017; Quezada-Euán et al. 2019). To determine whether genitalic parts are coordinating with the stabilizing selection prediction of strong negative allometry of genitalic structures, in Braconid wasps, this paper examined the allometry of different genitalic and non-genitalic morphological traits in males and females of *Habrobracon hebetor* (Say, 1836) (Hymenoptera: Braconidae). This species is a polyphagous and gregarious ectoparasitoid wasp which is widely used in integrated pest management programs. This species is mass-reared for control of the larvae of some important and well-known Lepidoptera pest species (Borzoui et al. 2016). The present paper also examined CV (coefficient of variation), CV´ (degree of dispersion of points around the allometric line) and SEE (standard error of estimate) that are other sources of information about the trait's variation. The results are discussed in evolutionary frameworks.

Material and Methods

Insects

A laboratory stock colony of *Habrobracon hebetor* was established from individuals collected from tomato fields during spring 2018 around Mashhad, Khorasan-e Razavi province of Iran. Rearing was done in the laboratory conditions at 25±1 ºC, 65±5 % RH, and a photoperiod of 16:8 (L:D) on fifth-stage larvae of *Ephestia kuehniella* for at least 10 generations for obtain to adequate population reared on the host at these conditions (Borzoui et al. 2016). 35 adult males and 35 adult females of *H. hebetor* were selected, randomly, from the laboratory stock colony and killed in Ethanol 70% solution.

E. kuehniella, were provided according to Borzoui et al. (2016) at the above-mentioned laboratory conditions. Examined materials were deposited in the Insect Collection of Birjand, Plant Protection Department, Faculty of Agriculture, University of Birjand, Iran.

Preparation and Measurements

Body parts of *H. hebetor* males and females were dissected. In each individual, the antenna, forewing, foreleg and hind leg were removed and cleaned to eliminate redundant tissues. Genitalia of both sexes were removed from the softened surrounding tissues of the abdomen and preserved in Ethanol 70% solution for only some minutes prior mounting. All mentioned parts were mounted on Canada balsam fixative. After preparation, all body parts were photographed through a microscope using a C-5050ZOOM digital camera (Olympus). 15 (three genitalic and 12 non-genitalic) traits in males and females were examined. For prevention of errors due to body asymmetry, only the right body parts were examined in all traits. The 12 measured non-genitalic traits in both sexes were the lengths of the following body parts: forewing, pterostigma, fore femur, fore tibia, spine of fore tibia, mid femur, mid tibia, hind femur, hind tibia, pedicel and the width of forewing and pterostigma (Fig. 1). The lengths of valva and ovipositor and the width of valva in the females and the lengths of the gonoforcep and penisvalva and the width of penisvalva in the males, were the measured genitalic traits (Fig. 1). TPSDIG, version 2.16 (Rohlf, 2015) was used for all the measurements. To minimize the measurement error, each trait was measured by the same person three times. Estimated measurement error for all studied traits showed relatively repeatable measurements (Table 1). Therefore, the average of the three measurements was used for further analyses.

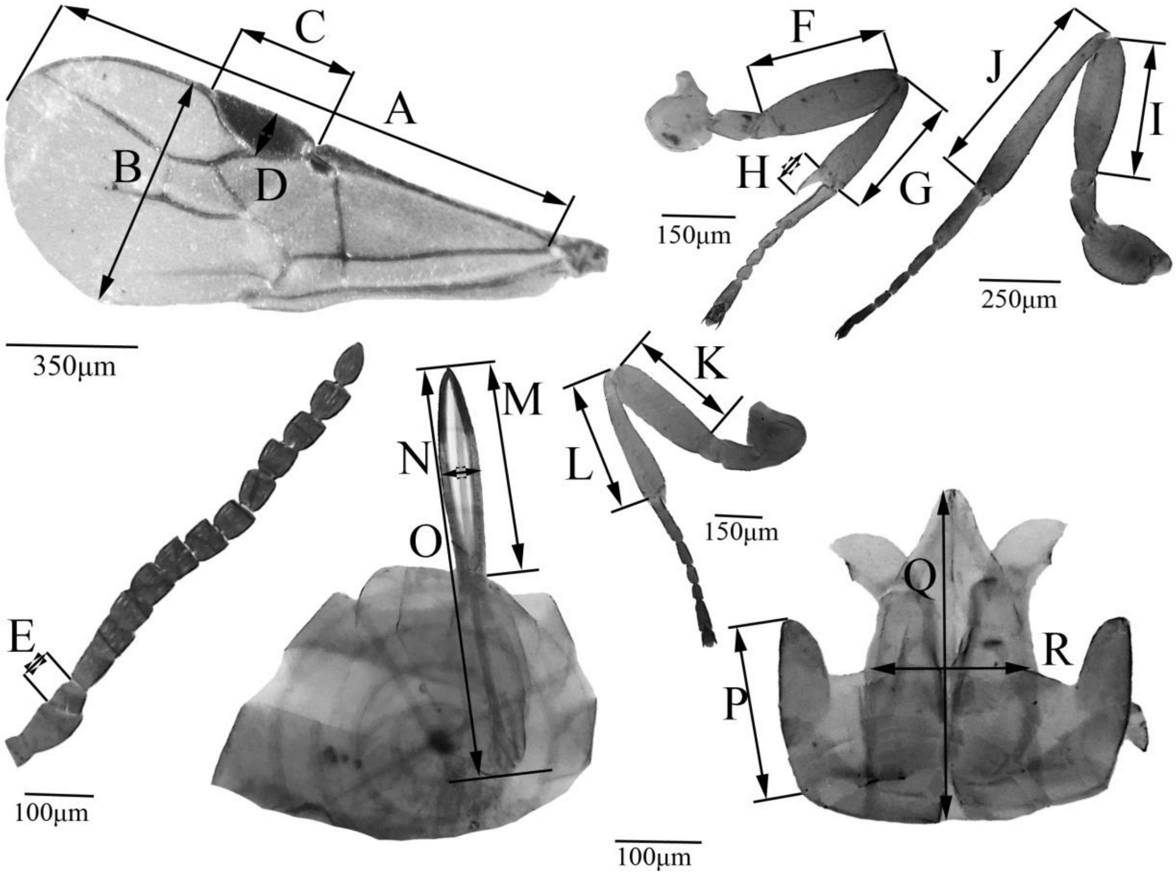

FIGURE 1. Genitalic and non-genitalic traits in male and female *Habrobracon hebetor*. A: Forewing length; B: Forewing width; C: Pterostigma length; D: Pterostigma width; E: Pedicel length; F: Fore femur length; G: Fore tibia length; H, Fore tibia spine length; I: Hind tibia length; J: Hind femur length; K: Mid femur length; L: Mid tibia length; M: Valva length; N: Valva width; O: Ovipositor length; P: Gonoforcep length; Q: Penisvalva length; R: Penisvalva width.

Data analysis

We used Principal Component Analysis (PCA), conducted separately for each sex, to obtain the best component explaining the variance of the traits. We selected forewing as the indicator of body size for allometry studies in male and female wasps. Loading all the measured traits on PC1, this trait showed the strongest correlation among other traits. Then, Pearson's product–moment correlation coefficient was calculated for the correlation between all other morphological traits and the body size indicator.

To estimate the allometric slopes, RMA and SMA regression analysis method was performed for log10-transformed values of the traits on log10-transformed forewing length in each sex according to Rabieh et al. (2015) (Table 2)

Also, CV (coefficient of variation), CV′ (degree of dispersion of points around the allometric line) and SEE (standard error of estimate) that are other sources of information about the trait's variation were calculated (Rabieh et al. 2015). The estimated components were compared between genitalic vs. non-genitalic traits by Mann–Whitney U-tests within each species. All statistical data analyses were performed using SPSS version 19.0 (IBM Corp, 2010).

TABLE 1. Measurement error and coefficients of variation (CV) evaluation for all male and female traits in *Habrobracon hebetor*

Trait		Measurement Error (%)	CV (%)
on-genitalic			
	Forewing length	0.78	2.11
	Forewing width	1.16	2.66
	Pterostigma length	1.25	1.93
	Pterostigma width	0.99	3.23
	Fore femur length	0.27	2.19
	Fore tibia length	0.82	1.5
	Fore tibia spine length	2.13	2.31
	Mid femur length	1.17	1.64
	Mid tibia length	1.4	2.47
	Hind femur length	1.55	2.43
	Hind tibia length	0.94	1.72
	Pedicel	2.21	2.13
Genitalic			
	Valva length	1.82	2.25
	Valva width	2.01	1.3
	Ovipositor length	1.96	2.32
	Penisvalva length	2.52	1.68
	Penisvalva width	2.71	1.21
	Gonoforcep length	2.3	1.12

TABLE 2. Allometry of evaluated morphometric traits in *Habrobracon hebetor* males.

Trait		r(p)	RMA slope	*p*-Value	SMA slope	SMA slope CI	CV′	SEE
Non-genitalic								
	Forewing width	0.92(0.0008)	0.88	< 0.001	0.85	0.74-1.02	1.04	0.0137
	Pterostigma length	0.88(0.0001)	0.83	< 0.001	0.77	0.67-1.00	0.92	0.0159
	Pterostigma width	0.89(0.005)	1.21	< 0.001	1.03	1.01-2.13	1.35	0.0217
	Fore femur length	0.85(0.0001)	0.67	< 0.001	0.89	0.52-1.01	1.15	0.0145
	Fore tibia length	0.86(0.004)	0.77	< 0.001	0.81	0.61-1.04	0.77	0.0167
	Fore tibia spine length	0.73(0.001)	0.78	< 0.001	0.73	0.52-1.03	1.26	0.0181
	Mid femur length	0.92(0.007)	0.74	< 0.001	0.65	0.63-1.22	0.64	0.0117
	Mid tibia length	0.87(0.0001)	0.75	< 0.001	0.82	0.6-1.16	1.22	0.0151
	Hind femur length	0.90(0.013)	0.84	< 0.001	0.79	0.7-1.31	1.06	0.0145
	Hind tibia length	0.83(0.012)	0.75	< 0.001	0.69	0.57-1.12	0.96	0.0178
	Pedicel	0.71(0.005)	0.89	< 0.001	0.81	0.57-1.21	1.50	0.0212
Genitalic								
	Penisvalva length	0.55(0.003)	0.2	0.006	0.18	0.06-0.34	1.04	0.0141
	Penisvalva width	0.42(0.006)	0.61	0.012	0.66	0.14-1.05	1.1	0.0157
	Gonoforcep length	0.53(0.05)	0.09	0.46	0.11	-0.16-0.34	0.94	0.0227

-The table shows Pearson correlation coefficient (r) and its *p*-value, ranged major regressions allometric coefficient (RMA slope), the *p*-value obtained from permutations to test if the allometric coefficients differed from zero, standardized major axis allometric coefficients (SMA slope), and their confidence intervals (SMA slope CI), degree of dispersion of points around the allometric line (CV′) and standard error of estimate (SEE).

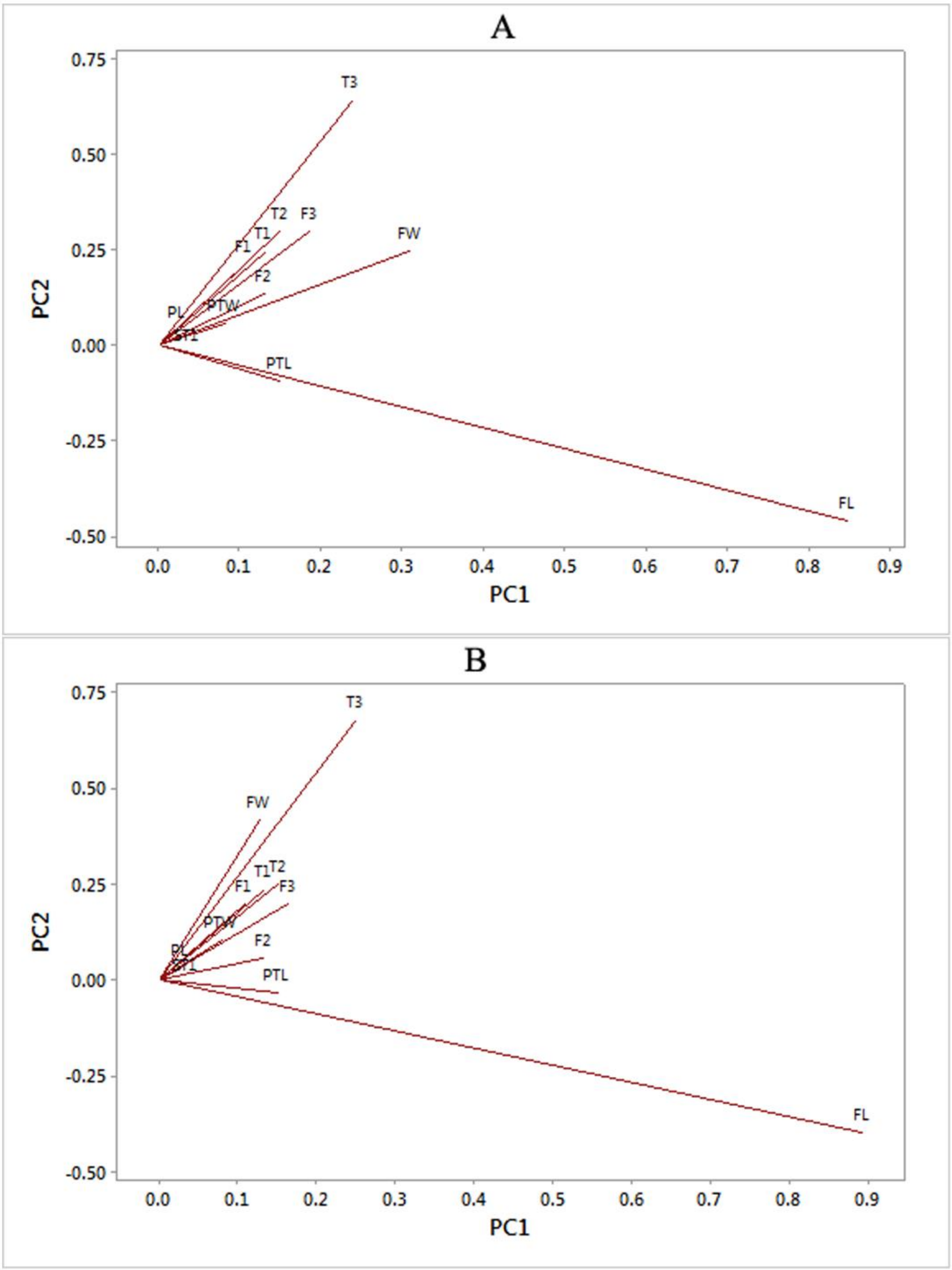

FIGURE 2. Principal component analysis (PCA) plots of trait loadings on PC1 vs. PC2 showing the covariation structure in the trait matrix of the males and females. A: *Habrobracon hebetor* male. B: *H. hebetor* female. FW: forewing width; FL: forewing length; F1: Fore femur length; F2: Mid femur length; F3: hind femur length; T1: Fore tibia length; T2: mid tibia length; T3: hind tibia length; ST1: Fore tibia spine length; PL; length of the pedicel; PTL: pterostigma length; PTW: pterostigma width.

TABLE 3. Allometry of evaluated morphometric traits in *Habrobracon hebetor* females.

Trait		r (p)	RMA slope	*p*-Value	SMA slope	SMA slope CI	CV′	SEE
Non-genitalic								
	Forewing width	0.94(0.0002)	0.84	< 0.001	0.89	0.72-1.00	0.91	0.0142
	Pterostigma length	0.89(0.0004)	0.81	< 0.001	0.79	0.71-1.01	0.88	0.0161
	Pterostigma width	0.84(0.001)	0.92	< 0.001	0.81	0.55-1.13	1.75	0.0188
	Fore femur length	0.79(0.0001)	0.61	< 0.001	0.69	0.63-1.05	1.34	0.0154
	Fore tibia length	0.81(0.002)	0.87	< 0.001	0.75	0.58-1.03	0.87	0.0173
	Fore tibia spine length	0.82(0.001)	0.6	< 0.001	0.68	0.57-1.01	1.32	0.0189
	Mid femur length	0.84(0.003)	0.53	< 0.001	0.52	0.43-1.00	0.88	0.0126
	Mid tibia length	0.88(0.0001)	0.68	< 0.001	0.83	0.62-1.12	1.17	0.0132
	Hind femur length	0.88(0.001)	0.82	< 0.001	0.77	0.7-1.25	1.15	0.0152
	Hind tibia length	0.9(0.002)	0.59	< 0.001	0.64	0.58-1.02	0.74	0.0138
	Pedicel	0.81(0.004)	0.79	< 0.001	0.85	0.67-1.19	1.2	0.0205
Genitalic								
	Valva length	0.48(0.05)	0.47	0.27	0.39	0.1-0.54	1.97	0.0311
	Valva width	0.69(0.01)	0.59	0.31	0.65	0.49-1.00	0.94	0.0251
	Ovipositor length	0.52(0.002)	0.62	0.01	0.8	0.53-1.01	1.98	0.0323

- The table shows Pearson correlation coefficient (r) and its *p*-value, ranged major regressions allometric coefficient (RMA slope), the *p*-value obtained from permutations to test if the allometric coefficients differed from zero, standardized major axis allometric coefficients (SMA slope), and their confidence intervals (SMA slope CI), degree of dispersion of points around the allometric line (CV′) and standard error of estimate (SEE).

DISCUSSION

This study is a comprehensive static allometry investigation on external genitalia and non-genitalic traits within both sexes of a Hymenopteran species. Although, most studies on insect's allometry usually focus on male genitalia traits and lay conspecific female genitalia away from their deduction.

In our results, almost all non-genitalia traits in both sexes of *Habrobracon hebetor* showed isometry (greater than 1.0 in pterostigma width in males). On the other hand, male genitalia traits of *H. hebetor* showed negative allometry except for Penisvalva width which showed isometry. This information confirms that, in *H. hebetor* male genitalia and non-genitalia traits reveal coordination with the one-size-fits-all hypothesis proposed by Eberhard et al. (1998) and further explained in Eberhard (2009). This means that in a given population of this species male individuals with average size of genitalia are more successful in generating surviving offspring than males with relatively smaller or larger genitalia sizes and this is irrespective of the overall body size of them. Our results in terms of the negative allometry of genitalia traits vs. isometry of non-genitalic traits, is in coordinate with the results of other works evaluated static allometry in male genitalia of different insect orders (e.g., Schmitz et al. 2000; Iwahashi, 2001; Tatsuta et al. 2001; Eberhard, 2002; Tschinkel et al. 2003; Hosken et al. 2005; Mutanen & Kaitala, 2006; Rabieh et al. 2015; Cao & Hayashi, 2019). Although, some studies found different results which do not support one-size-fits-all hypothesis, recently (e.g., Nava-Bolanos et al. 2012; Nava-Bolanos et al. 2014). These findings offer consideration on different functions of genitalia parts during copulation and sexual conflicts through different sexual selection mechanisms. For example, male genitalia parts may evolve to access stored sperm of females which leads to positive allometry of aedeagus in males of *Protoneura cara* (Odonata: Protoneuridae) (Nava-Bolanos et al. 2012).

According to our results, there is no significant difference between genitalic and non-genitalic traits in female wasps. This finding is another confirmation for the claim that the shallow static allometry slopes of male genitalic traits reflect functional roles associated with mating and sexual competition.

In a given species population, larger CV of a specified trait may be due to a higher allometric value or a greater dispersion of points around the allometric line (CV′). The allometric slope is related to genetic structure of the organism which evolves under different selection pressures but the degree of dispersion may be related not only to genetic structure but also to other environmental and internal causes (Eberhard, 1998). In our results, CV′ and SEE of genitalic traits have values less than those for non-genitalic traits, in the male wasps. Although, their difference was not significant, this result means that in *H. hebetor* the size of male genitalia is more stable than that of non-genitalic parts against changes in the body size, and the difference in the degree of phenotypic variation between genitalic and non-genitalic morphometric feature is related to the difference in allometric slopes. These findings are consistent with those of previous studies which investigated the allometric slope and the dispersion of points around the allometric line, separately, for male genitalia traits and other body parts (e.g., Eberhard et al. 1998; Ohno et al. 2003; Rabieh et al. 2015; Cao & Hayashi, 2019). It is assumed that sexual selection in arthropods may favor a particular scaling relationship for male genitalia under a developmental-genetic program which appears as allometric slope (Eberhard et al. 1998). Our results showed that such stabilizing sexual selection may also operate on genital size in braconid wasps.

Our results, presented another confirmation of the previous studies which showed that genitalia traits of females have lower allometric slopes than the median allometric slope for non-genitalic traits but this difference was not significant (e.g., Eberhard et al. 1998; Funke & Huber, 2005; Rabieh et al. 2015; Cao & Hayashi, 2019). We found that genitalia traits in *H. hebetor* females have lower allometric slope, CV, CV′ and SEE than those for non-genitalic traits which for two last components, the differences were significant. Our findings showed similar allometric patterns among the male and female traits measured. Consequently, as male genitalia parts showed negative allometry and female genitalia, on the other hand displayed isometric relationship, we predict relatively invariability in female morphology of *H. hebetor* so that male wasp could adapt to this by being consistent in genital morphology. While our evidences do suggest the possibility of stabilizing selection in *H. hebetor*, additional experiments would be required to reinforce these claims.

If a non-genitalic trait of a male individual attends as a secondary sexual trait during sexual selection mechanism, it may produce higher mating success when has a larger relative size, conversely, in small individuals, viability costs may limit the size of this trait, resulting in positive allometry (Bonduriansky, 2007). This will be much distinctive when a large number of competitive males exist or the trait produces resolutely more advantage in competitive interactions (Fromhage & Kokko, 2014). In our results, the pterostigma width in *H. hebetor* males showed positive allometry in relation to body size. Conversely, this trait in *H. hebetor* females showed an isometric relationship with body size. This suggests selection for larger pterostigma in large *H. hebetor* males and/or relatively smaller ones in small *H. hebetor* males. Therefore, positive allometry of the pterostigma width in *H. hebetor* may be a result of secondary sexual function. This possibility deserves further investigations.

RESULTS

The first and second components of the PCA explained 94.8% and 3.4% of the total variance in the males and 91.1% and 3.7% in the females, respectively. Therefore, PC1 chose as the best component explaining the variance of the traits for both sexes. Plots of trait loadings on PC1 vs. PC2 showing the covariation structure in the trait matrix of the males and females are presented (Fig. 2). Loading all the measured traits on PC1, forewing length showed the strongest correlation among other traits (Males: r = 0.989, *p*-value < 0.001; Females: r = 0.977, *p*-value < 0.001). All the measured traits were significantly related to the overall body size for male and female wasps

(Tables 2 and 3). In most traits, a highly significant relationship (*p*-value<0.01) was observed in both sexes (Tables 2 and 3). All the non-genitalic traits in male and female wasps showed isometric relationship with the body size indicator (Tables 2 and 3) except pterostigma width in male wasps which showed positive allometric relationship with body size (Table 2). In females, valva length showed strongly negative allometric relationship with body size and valva width and ovipositor length showed isometry (Tables 3). In male wasps, length of penisvalva and gonoforcep have strong negative allometry in relation to body size indicator and penisvalva width showed values which did not differ significantly from one (isometry) (Table 2).

The mean slope for the measured genitalic traits of males was significantly lower than one's for the non-genitalic traits (Mann–Whitney U-test, $P < 0.01$). In female wasps, there is no significant difference (Mann–Whitney U-test, $P = 0.097$) but the mean slope of genital traits was lower than the mean slope for non-genitalic traits. There is no significant difference between results of allometric slopes in the RMA and SMA regression methods (Mann–Whitney U-test, $P < 0.01$).

Genitalic traits of male wasps showed significantly lower CV than non-genitalic traits (Mann–Whitney U-test, $P < 0.01$). Despite the lower mean value for genitalic traits, there was no significant difference between CV for genitalic and non-genitalic traits, in female wasps (Tables 3). Conversely, CV′ showed no significant difference between genitalic and non-genitalic traits in male wasps (Mann–Whitney U-test, $P = 0.361$) and it was significantly lower for non-genitalic than genitalic traits in female wasps. Similar situation was seen for SEE which only female wasps have significantly lower non-genitalic traits than genitalic traits (Tables 2 and 3).

Acknowledgments

We sincerely thank Dr. Javad Noei for his kind helps in the laboratory affairs. This study was supported by the grant, number 21833, from university of Birjand, Birjand, Iran.

LITERATURE CITED

Andersson, M., 1994. Sexual Selection. Princeton University Press, Chichester, UK.

Benítez, H.A., Bravi, R., Parra, L.E., Sanzana, M-J., Sepúlveda-Zúñiga, E., 2013. Allometric and non-allometric patterns in sexual dimorphism discrimination of wing shape in *Ophion intricatus*: Might two male morphotypes coexist? Journal of Insect Science 13, 143.

Bonduriansky, R., 2007. Sexual selection and allometry: a critical reappraisal of the evidence and ideas. Evolution 61, 838–849.

Boulton, R.A., Collins, L.A., Shuker, D.M., 2015. Beyond sex allocation: the role of mating systems in sexual selection in parasitoid wasps. Biological Reviews of the Cambridge Philosophical Society 90, 599–627.

Borzoui, E., Naseri, B., Mohammadzadeh-Bidarani, M., 2016. Adaptation of *Habrobracon hebetor* (Hymenoptera: Braconidae) to rearing on *Ephestia kuehniella* (Lepidoptera: Pyralidae) and *Helicoverpa armigera* (Lepidoptera: Noctuidae). Journal of insect science 16, 1–12.

Cao, C., Yu, P., Hayashi, F., 2019. Allometry and morphological trait relationship in the sexually dimorphic Chinese dobsonfly, *Acanthacorydalis asiatica* (Wood-Mason, 1884) (Megaloptera, Corydalidae). ZooKeys 854, 119–129.

Cayetano, L., Maklakov, A.A., Brooks, R.C., Bonduriansky, R., 2011. Evolution of male and female genitalia following release from sexual selection. Evolution 65, 2171–2183.

Eberhard, W.G., 1985. Sexual selection and animal genitalia. Harvard University Press. Cambridge, UK.

Eberhard, W.G., 2002. The relation between aggressive and sexual behavior and allometry in *Palaeosepsis dentatiformis* flies (Diptera: Sepsidae). Journal of the Kansas Entomological Society 75, 317–332.

Eberhard, W.G., 2009. Static allometry and animal genitalia. Evolution 63, 48–66.

Eberhard, W.G., Huber, B.A., Rodriguez, R.L., Briceno, R.D., Salas, I., Rodriguez, V., 1998. One size fits all? Relationships between the size and degree of variation in genitalia and other body parts in twenty species of insects and spiders. Evolution 52, 415–431.

Fromhage, L., Kokko, H., 2014. Sexually selected traits evolve positive allometry when some matings occur irrespective of the trait. Evolution 68(5), 1332–1338.

Funke, S., Huber, B.A., 2005. Allometry of genitalia and fighting structures in *Linyphia Triangularis* (Araneae, Linyphiidae). The Journal of Arachnology 33, 870–872.

Green AJ. 1999. Allometry of genitalia in insects and spiders: one size does not fit all. Evolution 53: 1621–1624.

Hosken, D.J., Stockley, P., 2004. Sexual selection and genital evolution. Trends in Ecology and Evolution 19, 87–93.

Hosken, D.J., Minder, A.M., Ward, P.I., 2005. Male genital allometry in Scathophagidae (Diptera). Evolutionary Ecology 19(5), 501–515.

Iwahashi, O., 2001. Aedeagal length of the Oriental fruit fly, *Bactrocera dorsalis* (Hendel) (Diptera: Tephritidae), and its sympatric species in Thailand and the evolution of a longer and shorter aedeagus in the parapatric species of *B. dorsalis*. Applied Entomology and Zoology 36, 289–297.

International Business Machines Corporation, 2010. IBM SPSS Statistics for Windows, Version 19.0. Armonk, NY.

Johnson, N.F., 1995. Variation in male genitalia of Merope tuber Newman Mecoptera: Meropeidae. Journal of the Kansas Entomological Society 682, 224–233.

Leonard, J., Cordoba-Aguilar, A., 2010. The evolution of primary sexual characters in animals. Oxford University Press, NY.

Mutanen, M., Kaitala, A., 2006. Genital variation in a dimorphic moth *Selenia tetralunaria* (Lepidoptera, Geometridae). Biological Journal of the Linnean Society 87, 297–307.

Nava-Bolanos, A., Mungua-Steyer, R., Cordoba-Aguilar, A., 2012. A test of genital allometry using two damselfly species does not produce hypoallometric patterns. Ethology 118, 203–213.

Nava-Bolanos, A., Sanchez-Guillen, R.A., Mungua-Steyer, R., Cordoba-Aguilar, A., 2014 Isometric patterns for male genital allometry in four damselfly species. Acta Ethologica 17, 47–52.

Ohno, S., Hoshizaki, S., Ishikawa, Y., Tatsuki, S., Akimoto, S., 2003. Allometry of male genitalia in a Lepidopteran species, *Ostrinia latipennis* (Lepidoptera: Crambidae). Applied Entomology and Zoology
38: 313–319.

Perrard, A., Villemant, C., Carpenter, J.M., Baylac, M. 2012. Differences in caste dimorphism among three hornet species (Hymenoptera: Vespidae): forewing size, shape and allometry. Journal of Evolutionary Biology 25, 1389–1398.

Polilov, A.A., Makarova, A.A., 2017. The scaling and allometry of organ size associated with miniaturization in insects: A case study for Coleoptera and Hymenoptera. Scientific Reports 7, 43095.

Quezada-Euán, J.G., Corey, S., Raúl, S., Castillo, C., 2019. Patterns of sexual size dimorphism in stingless bees: Testing Rensch's rule and potential causes in highly eusocial bees (Hymenoptera: Apidae, Meliponini). Ecology and Evolution 9, 2688–2698.

Rabieh, M.M., Esfandiari, M., Seraj, A.A., Bonduriansky, R., 2015. Genital and body allometry in two species of noctuid moths (Lepidoptera: Noctuidae). Biological Journal of the Linnean Society 116, 183–196.

Rohlf, F.J. 2015. TPSDIG, version 2.16. Department of Anthropology, Stony Brook University, Stony Brook, USA, Available online at: http://life.bio.sunysb.edu/morph/.

Schmitz, G., Reinhold, K., Wagner, P., 2000. Allometric relationship between genitalic size and body size in two species of mordellid beetles (Coleoptera: Mordellidae). Annals of the Entomological Society of America 9(3), 637–639.

Tatsuta, H., Mizota, K., Akimoto, S.I., 2001. Allometric patterns of heads and genitalia in the stag beetle *Lucanus maculifemoratus* (Coleoptera: Lucanidae). Annals of the Entomological Society of America 94, 462–466.

Tschinkel, W.R., Mikheyev, A.S., Storz, S.R., 2003. Allometry of workers of the fire ant, *Solenopsis invicta*. 11pp. Journal of Insect Science 3, 2.

West, S.A., 2009. Sex allocation. Princeton University Press, Woodstock, UK.