J. Entomol. Res. Soc., 23(3): 213-224, 2021
Doi: 10.51963/jers.v23i3.2010

Research Article
Online ISSN:2651-3579

Differences Between the Allometric Rules Governing Two *Formica lugubris* Zetterstedt, 1838 (Hymenoptera: Formicidae) Ants Populations in Northern Greece

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ABSTRACT

Red wood ant workers, despite being monomorphic, show a wide range in their body size even within the same colony. As overall size grows, some organs grow in a negative allometric relationship with the body. Genetic and environmental factors, like diet and temperature, are considered as key factors that determine differences in body and organ growth. We studied the allometric rules governing the organs in two different populations of red wood ants in two different environments in Northern Greece to identify similarities and differences. These rules, although linear for both populations and negative allometric for most of the traits measured, differ in the two study areas, while the shape of the head found to be governed by different allometric rules between the two populations. The whole shape and size of red wood ants varies greatly even within the same species and we cannot create allometric models for two different areas, even when they belong in the same species. The differences might be a result of differences in the genome and the environmental conditions between the two populations.

Key words: Red wood ants, *Formica lugubris*, morphological traits, allometry, Northern Greece.

Tsikas, A. & Karanikola, P. (2021). Differences between the allometric rules governing two *Formica lugubris* Zetterstedt, 1838 (Hymenoptera: Formicidae) ants populations in northern Greece. *Journal of the Entomological Research Society*, 23(3), 213-224.

INTRODUCTION

Body size is the main phenotypic feature of organisms, as it affects the way they interact with the environment. In nature, adult individuals of the same species may vary widely in body size due to a combination of genetic and environmental factors. This is the result of differential growth: as an individual organism grows in size overall, some organs grow faster or slower than others (Eberhard et al, 1998; Emlen, Warren, Johns, Dworkin, & Lavine, 2012) due to differential allocation of resources to different organs, at different rates (Bonduriansky, Day, & Pitnick, 2003; Kodric-Brown, Sibly, & Brown, 2006; Emlen et al, 2012).

Variations in the size of an organ that accompany variations in body size can be characterized by allometric relationships. The scaling of any quantitative characteristic of an organ can be modelled using the allometric equation,

$y=bx^a$

Where x is the size of the body, Y is the size of the instrument, a is the exponent of scaling and b is the initial growth index (Huxley & Tessier, 1936). A log transformation of the allometric equation produces a simple linear equation, $\log(y)=\log(b)+a \log(x)$ and log-log plots of the size of different traits among individuals of the same species typically reveal linear allometries with an intercept of log(b) and a slope of a, called the ‘allometric coefficient’ (Huxley & Tessier, 1936). When the organs grow in the same rate with the rest of the body ($a = 1$) then we have isometry. More commonly, however, the organs have a negative allometric relationship with the body ($a < 1$), *i.e.* they grow at a relatively slower rate than the body, resulting in larger individuals having relatively smaller organs (Eberhard et al, 1998), while in rare cases, and especially in organs related to sexual choice, the organs may show positive allometric relationships ($a > 1$), *i.e*. they grow in a faster rate than the body (Gould, 1973; Emlen, 1997). In some cases, however, the equation is not always linear, and may be sigmoidal or intermittent (Emlen & Nijhout, 2000).

In holometabolous insects, the organs do not grow at the same time as the larva, but in the later stages of larval development from extracellular monolayers called imaginal discs (Morata & Lawrence, 1979; Currie, Milner, & Evans, 1988). The differential resource allocation in the imaginal discs is resolved through insulin-like peptides and their receptors, making nutrition the most important factor for size, and consequently for scaling (Shingleton, Das, Vinicius, & Stern, 2005). Hence, the nutritional quality of larval diets impacts the metabolic functioning of imago insects, with diets more optimal for survival resulting in a higher metabolic rate per unit of body mass (Nicholls, Rossi, & Niven, 2021). In addition, because larvae stop feeding before metamorphosis, imaginal disc development takes place under conditions of constant resources in the form of protein and fat storage (Tschinkel, 2013). Therefore, there is competition among imaginal discs for limited resources, and the increase of one is accompanied by the decrease of another (Nijhout & Wheeler, 1996). Other factors that determine differences in the size and scaling rules of insect organs are the genome (Stevenson, Hill, & Bryant, 1995; Bargum, Boomsma, & Sundström, 2004; Shingleton, Estep, Driscoll, & Dworkin, 2009) and temperature (Shingleton et al, 2009;

Nijhout et al, 2014). Therefore, it is expected that allometric rules in holometabolous insects will be different from those that apply to other animals whose organs develop continuously and simultaneously with their overall body.

The peculiarity of the development of imago holometabolous insects has two consequences on allometry (Nijhout & Wheeler, 1996). First, where allomorphic relations are observed between organs and the whole body, they cannot be interpreted as a mere result of relative growth, as the organs develop at different times and under completely different regimes of endocrine and nutritional conditions compared to the body. Second, some rare cases of allometric relationships, such as sigmoid and intermittent allometry observed in some polymorphic ant worker castes (Feener, Lighton, & Bartholomew, 1988; Wheeler, 1991) but also in some beetles (Emlen, 1994; Kawano, 1995), may be related to interaction of imaginal discs with each other during metamorphosis.

Ants are organisms with large intra-specific variation in body size (Kaspari & Weiser, 1999; Geraghty, Dunn, & Sanders, 2007; Hurlbert, Ballantyne, & Powell, 2008). The size variation in ants, because they are holometabolous insects, is determined mostly by the conditions of their growth, and mainly by the trophic regime of the larvae. As food is distributed by the workers to the larvae, a higher ratio of the number of workers to that of the larvae means that the larvae receive more resources (Fedoseeva, 2011). The trophic regime is affected by the abundance of prey, as well as by certain factors that suppress the food of the workers or increase their mortality. One year after clear-cutting, Sorvari & Hakkarainen (2009) found that the workers from *F. aquilonia* nests in the deforested areas were smaller in size than in undisturbed stands.

Red wood ants (*Formica rufa* Group) workers are monomorphic and not divided into different morphological castes, but show a wide range in their body size, and even within the same colony (Perl & Niven, 2016) with workers working outside the nest being smaller than workers on the nest (Véle & Mondlinger, 2019). The purpose of this work was to study the allometric rules governing the organs in two different populations of red wood ants in two different environments, with different soil, climatic and vegetation conditions, and their comparison to identify similarities and differences. Given the monomorphism, we expected to find linear allometric relations between their organs and their body.

MATERIAL AND METHODS

Study area

The study areas are located in Elatia Forest, Drama (41° 29′ N, 24° 18′ E), and in Lailias Forest, Serres (41° 14’ N, 23° 34’ E), both in Northern Greece. The altitude of the study area in Elatia ranges between 1500-1650 meters with a mild to moderately strong slope, while the altitude of the study area in Lailias ranges between 1450-1550 meters with a mild to moderate strong slope. They are both natural managed mixed forests, where Elatia is dominated by spruce (*Picea abies* L.), while Lailias is dominated by Scots pine (*Pinus sylvestris* L.). The geological formation in Elatia consists mainly of granite, gneiss, and limestone (Zagas, 1990), and in Lailias is dominated by acidic igneous rocks (granites, granodiorites, monzonites) with lake and soil reserves in the lowlands (Flocas, Giles, & Angouridakis, 1983).

Sampling and measurements

We collected manually and randomly from the top of the nest mound individual ants from 30 mounds from Elatia in 2013, and from 30 mounds from Lailias in 2017. Samples were taken in summer in the daytime. All samples were preserved in ethanol in glass vials. The species were identified by the keys of Agosti & Collingwood (1987) and Stockan, Robinson, Trager, Yao, & Seifert (2016). 5 individuals were sampled from these vials; a total of 300 individuals. They were then cut with a scalpel, and the pieces (head, antennae, legs, mesosoma) were arranged on a gridded, numbered card that was covered with double-stick tapes to hold them in place. The head was arranged in a face-on view and the mesosoma in lateral view. The pieces were photographed on the stereoscope OLYMPUS SZX7 with a NIKON D90 camera at 10x magnification and with the help of a scale their morphological characteristics were measured with the program Natsumushi v 1.10.1.

The measured traits were: head width (HW), head length (HL), the length of the scape (SL), the length of the upper jaw (MandL), the length of the mesosoma or Weber's length (WL), and the hind femur length (HFL). The head index (CI = 100 * HW / HL), the scape index (SI = 100 * SL / HW) and the mandible index (MandIndex = 100 * MandL / HL) were then calculated. The explanation of the abbreviations is given in Table 1.

Table 1. Selected morphological traits measured in workers.

Abbreviation	Description	Metric
HW	The maximum width of the head in full face view	mm
HL	The length of the head capsule excluding the mandibles, measured in full face view in a straight line from the mid-point of the anterior clypeal margin to the mid-point of the posterior margin	mm
SL	The maximum straight-line length of the scape	mm
MandL	The straight-line length from the mandibular apex to the anterior clypeal margin	mm
WL	The diagonal length of the mesosoma in profile from the point at which the pronotum meets the cervical shield to the posterior basal angle of the metapleuron	mm
HFL	Maximum length of hind femur, measured in anterior view	mm

Data analysis

Standard deviation (± SD) and the coefficient of variation (CV%) were calculated for all measured traits. HFL was used as an indicator of body size, and allometric relationships with the other traits were calculated after log-log transformation. The slope of the regression was calculated by the regression of the maximum axis, as the least squares method tends to produce devaluations (McArdle, 1988). When two measured traits change accordingly, the slope of the regression is equal to 1 (Zar, 1999). When the regression slope = 1, we have an isometric relation. A slope < 1 indicates negative allometry, ie the organs grow at a slower rate compared to the overall size, while when the slope > 1 we have positive allometry. Then we made an estimate of the shape of the head independent of its size with the log-log ratio of the two dimensions (Log10 HW / HL) to log of the index of the total size, HFL (Mosiman & James, 1979). When the slope does not differ statistically significantly from zero (t-test) it shows us that the shape of the head does not change with increasing body size, ie it is isometric with the body, while a slope other than zero shows an allometric relationship of the shape with the overall size. The distribution of all measurements was found to be normal with homogeneous variations. IBM SPSS Statistics v.23 was used for the analyses and Past v2.17c was used for the graphs.

RESULTS

All nests in both study areas were found to belong to *Formica lugubris* Zetterstedt, 1838. The results of the measurements of 150 individuals in Elatia and 150 individuals in Lailias, as well as the standard deviation and the coefficient of variation are presented in Table 2. In general, no large morphological variations were found in the measured traits or significant statistical differences, but the samples from Lailias were found to be slightly larger. Little variation was found in the head, scape and mandible indexes (Table 3).

Table 2. Measured traits in *F. lugubris* workers in Elatia and Lailias.

Elatia						
	HFL (mm)	**HW (mm)**	**HL (mm)**	**SL (mm)**	**MandL(mm)**	**WL (mm)**
N	150	150	150	150	150	150
Mean	1,96102	1,591993	1,222197	1,490527	0,817434	2,44576
SD	0,2249747	0,1728537	0,1267583	0,1587119	0,083282	0,25175
CV %	11,47233	10,85769	10,37135	10,64804	10,18822	10,2934
Lailias						
	HFL (mm)	**HW (mm)**	**HL (mm)**	**SL (mm)**	**MandL(mm)**	**WL (mm)**
N	150	150	150	150	150	150
Mean	2,157107	1,66512	1,262226	1,631273	0,8471	2,61252
SD	0,182989	0,138613	0,108357	0,111026	0,066746	0,20873
CV %	8,483065	8,324524	8,584612	6,80607	7,879305	7,98942

HW found linearly related to HFL (and consequently to the total body size both in Elatia ($R^2 = 0.693$, $p < 0.0001$, log-log slope = 0.783) (Fig. 1a), and in Lailias ($R^2 = 0.747$, $p < 0.0001$, log-log slope = 0.853) (Fig. 2a). Respectively, HL is related to HFL in Elatia ($R^2 = 0.587$, $p < 0.0001$, log-log slope = 0.678) (Fig. 1b) and in Lailias ($R^2 = 0.707$, $p < 0.0001$, log-log slope = 0.85) (Fig. 2b). SL grows at slower rate than to the overall size, as does the length of the Mandible. For Elatia, for SL ($R^2 = 0.714$,

$p < 0.0001$, log-log slope = 0.78) (Fig. 1c), while for MandL ($R^2 = 0.563$, $p < 0.0001$, log-log slope = 0.684) (Fig. 1d). For Lailias, for SL ($R^2 = 0.684$, $p < 0.0001$, log-log slope = 0.662) (Fig. 2c), while for MandL ($R^2 = 0.699$, $p < 0.0001$, log-log slope =0.771) (Fig. 2d). Finally, WL found to have negative allometry with body size in both Elatia ($R^2 = 0.69$, $p < 0.0001$, log-log slope = 0.736) (Fig. 1e), and in Lailias ($R^2 = 0.788$, $p < 0, 0001$, log-log slope = 0.831) (Fig 2e).

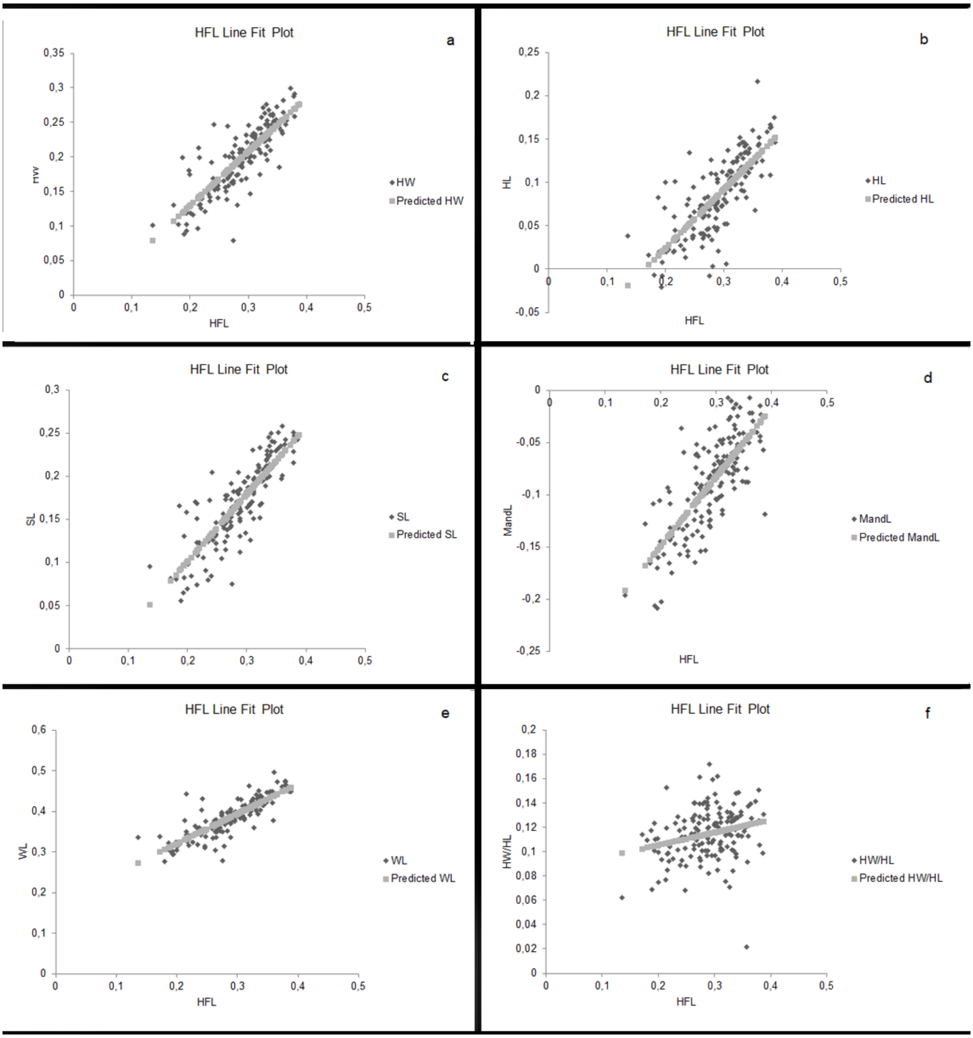

Fig 1. The relationship between HFL and a) HW (y = 0,783x – 0,027), b) HL (y= 0,678x – 0,112), c) SL (y = 0,78x – 0,055), d) MandL (y = 0,684x - 0,282), e) WL (y = 0,736x + 0,173), and f) Head Ratio (y = 0,105x + 0,084) in Elatia.

Despite the allometric rules in both populations in all measured traits, there is a difference in the head ratio, and consequently the shape of the head. Thus, for Elatia we found the shape of the head capsule to have an allometric relationship with the body (R^2 = 0.061, p = 0.002264, log-log slope = 0.105) (Fig. 1f), while for Lailias the shape of the head does not change with increase in body size and has an isometric relationship with the body (R2 = 0.0001, p = 0.9, log-log slope = 0.003) (Fig. 2f).

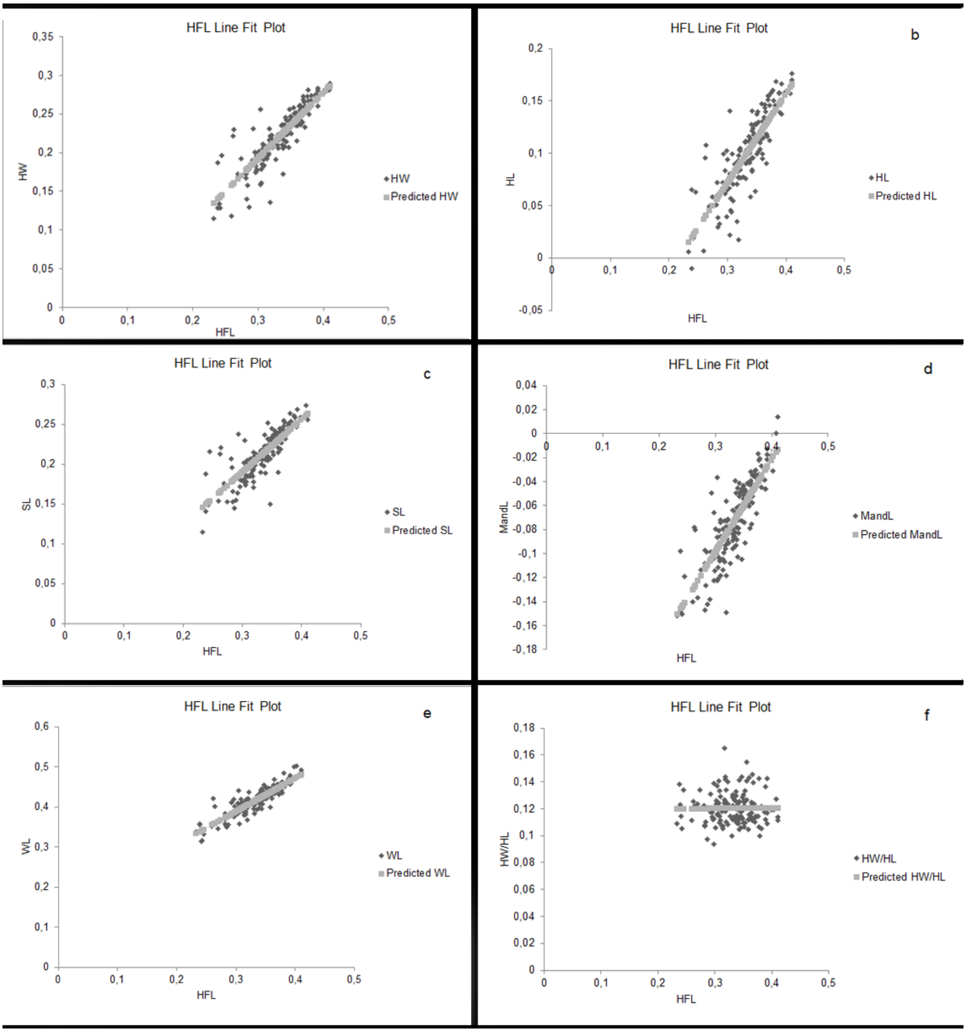

Fig 2. The relationship between HFL and a) HW (y = 0,853x – 0,064), b) HL (y = 0,85x – 0,183), c) SL (y = 0,662x – 0,008), d) MandL (y = 0,771x - 0,330), e) WL (y = 0,831x + 0,140), and f) Head Ratio (y = 0,003x + 0,119) in Lailias.

Table 3. Cephalic Index (CI), Scape Index (SI) and Mandible Index (MandIndex) in *F. lugubris* workers in Elatia and Lailias.

Elatia			
	CI	**SI**	**MandIndex**
N	150	150	150
Mean	130,332	93,8729	67,1004
SD	6,38762	6,55454	5,364791
CV %	4,90103	6,98236	7,995169
Lailias			
	CI	**SI**	**MandIndex**
N	150	150	150
Mean	124,013	98,1469	60,10378
SD	3,58943	3,50959	2,633692
CV %	2,71943	3,57586	3,91895

DISCUSSION

Red wood ant workers, although monomorphic and not divided into castes, show a wide range of size in their body. Using the length of the femur of the hind leg as a measure of size, we created models with allometric rules governing the size of the workers' organs. These rules, although linear for both populations, differ in the two study areas.

As the size of the worker's body increases, so does the size of the head, the scapi, the mandible and the mesosoma, but with negative allometry. Consequently, smaller workers have relatively larger organs compared to larger ones. These relationships occur in all nests we studied in both study areas. Seifert (2016) found similar results for *F. lugubris*, while Perl, Rossoni, & Niven (2017) found grade shifts in the allometric scaling of the compound eye among four *Formica* species in terms of eye scaling, but no grade or slope shifts in the scaling of mean facet diameter. Also, similar relationships have been observed in other ants such as e.g. *Melophorus bagoti* (Schwarz et al, 2011), as well as in other Hymenoptera, such as *Bombus terrestris* (Spaethe & Chittka, 2003). However, the comparison between the population of Elatia and Lailias, shows us significant differences in the allometric rules between the populations. Consequently, we cannot create allometric models for red wood ants from two different areas, even when they belong in the same species. Also, regarding the shape of the head, it is observed that in Elatia it also has allometric relations with the size, while on the contrary in Lailias it does not seem to change. Therefore, the whole shape and size of red wood ants seems to vary greatly even within the same species. Future research should focus on the factors that lead us to these results. A genetic analysis of the two populations may have shown us possible differences in their genome. The genetic diversity is expected to be low in these isolated populations, due to the genetic bottleneck. So, the two populations are expected to differ significantly due to zero gene flow, or even it is possible to have two different species. Molecular analyses have been used for the identification and monitoring of cryptic species (e.g. Bickford et al, 2007; Vogler &Monaghan, 2007; Moreau, 2009). Morphologically similar ant species have been found to differ significantly in mtDNA analyses (Smith, Fisher, & Hebert, 2005; Steiner et al, 2005; Pusch, Seifert, Foitzik, & Heinze, 2006; Schlick-Steiner et al, 2006a; 2006b; Steiner et al, 2006; Bernasconi, Pamilo, & Cherix, 2010) or microsatellites (Macaranas, Colgan, Major, Cassis, & Gray, 2001; Gyllenstrand, Seppä, Pamilo, 2004; Knaden, Tinaut, Cerda, Wehner, & Wehner, 2005; Bernasconi et al, 2010).

Also, the two populations have different environmental conditions, such as vegetation and temperature, and consequently diet, which could explain the differences in allometric relations. Larvae diet (Thomas, 1993) and temperature (Atkinson, 1994; Mirth & Riddiford, 2007) directly affect adult size. Both diet and temperature were found to affect allometric relationships in the fly *Drosophila melanogaster* (Shingleton et al, 2009), which is also holometabolous, like red wood ants. The various factors that affect growth, like the differences in scots pine and spruce-sucking aphids' honeydew nutrients, should be studied in the future in different ecosystems for possible correlations.

REFERENCES

Agosti, D. & Collingwood, C.A. (1987). A provisional list of the Balkan ants (Hym. Formicidae) with a key to the worker caste. II. Key to the worker caste, including the European species without the Iberian. *Mitteilungen der Schweizerischen Entomologischen Gesellschaft*, 60, 261-293.

Atkinson, D. (1994). Temperature and organism size - A biological law for ectotherms? *Advances in Ecological Research*, 25, 1-58.

Bargum, K., Boomsma, J.J., & Sundström, L. (2004). A genetic component to size in queens of the ant *Formica truncorum*. *Behavioral Ecology and Sociobiology*, 57, 9-16.

Bernasconi, C., Pamilo, P., & Cherix, D. (2010) Molecular markers allow sibling species identification in red wood ants (*Formica rufa* group). *Systematic Entomology*, 35, 243-249.

Bickford, D., Lohman, D.J., Sohdi, N.S., Ng, P.K.L., Meier, R., Winker, K., Ingram, K. & Das, I. (2007). Cryptic species as a window on diversity and conservation. *Trends in Ecology & Evolution*, 22, 148-155.

Bonduriansky, R., Day, T., & Pitnick, S. (2003). The evolution of static allometry in sexually selected traits. *Evolution,* 57, 2450-2458.

Currie, D.A., Milner, M.J., & Evans, C.W. (1988). The growth and differentiation in vitro of leg and wing imaginal disc cells from *Drosophila melanogaster*. *Development*, 102, 805-814.

Eberhard, W.G., Huber, B.A., Rodriguez R.L.S., Briceño, 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(2), 415-431.

Emlen, D.J. (1994). Environmental control of horn length dimorphism in the beetle *Onthophagus acuminatus* (Coleoptera: Scarabaeidae). *Proceedings of the Royal Society of London. Series B, Biological Sciences*, 256, 131-136.

Emlen, D.J. (1997). Diet alters male horn allometry in the beetle *Onthophagus acuminatus* (Coleoptera: Scarabaeidae). *Proceedings of the Royal Society of London. Series B*, 264, 567-574.

Emlen, D.J. & Nijhout, H.F. (2000). The development and evolution of exaggerated morphologies in insects. *Annual Review of Entomology*, 45, 661-708.

Emlen, D.J., Warren, I.A., Johns, A., Dworkin, I., & Lavine, L.C. (2012). A mechanism of extreme growth and reliable signaling in sexually selected ornaments and weapons. *Science*, 337(6096), 860-4.

Fedoseeva, E.F. (2011). Morphometric characteristics of *Formica aquilonia* ants in monitoring of their settlements. *Entomological Review*, 91(2), 152-168.

Feener, D.H, Lighton, J.J.R.B., & Bartholomew, G.A. (1988). Curvilinear allometry, energetics and foraging ecology: A comparison of leaf-cutting ants and army ants. *Functional Ecology*, 2(4), 509-520.

Flocas, A.A., Giles, B.D., & Angouridakis, V.E. (1983). On the estimation of annual and monthly mean-values of air-temperature over Greece using stepwise multiple-regression analysis. *Archives for Meteorology Geophysics and Bioclimatology Series B-Theoretical and Applied Climatology*, 32(2-3), 287-295.

Geraghty, M.J., Dunn, R.R., & Sanders, N.J. (2007). Body size, colony size, and range size in ants (Hymenoptera: Formicidae): are patterns along elevational and latitudinal gradients consistent with Bergmann's Rule? *Myrmecological News*, 10, 51-58.

Gould, S.J. (1973). Positive allometry of antlers in the 'Irish Elk' *Megaloceros giganteus*. *Nature*, 244, 375-376.

Gyllenstrand, N., Seppä, P., & Pamilo, P. (2004). Genetic differentiation in sympatric wood ants, *Formica rufa* and *F. polyctena*. *Insectes Sociaux*, 51, 139-145.

Hurlbert, A.H., Ballantyne I.V.F., & Powell, S. (2008). Shaking a leg and hot to trot: the effects of body size and temperature and running speed in ants. *Ecological Entomology*, 33, 144-154.

Huxley, J.S. & Tessier, G. (1936). Terminology of relative growth. *Nature*, 137, 780-787.

Kaspari, M. & Weiser, M.D. (1999). The size-grain hypothesis and interspecific scaling in ants. *Functional Ecology*, 13, 530-538.

Kawano, K. (1995). Horn and wing allometry and male dimorphism in giant Rhinoceros beetles (Coleoptera: Scarabaeidae) of tropical Asia and America. *Annals of the Entomological Society of America*, 88(1), 92-99.

Knaden, M., Tinaut, A., Cerda, X., Wehner, S., & Wehner, R. (2005). Phylogeny of three parapatric species of desert ants, *Cataglyphis bicolor*, *C. viatica* and *C. savigny*: a comparison of mitochondrial DNA, nuclear DNA and morphological data. *Zoology*, 108, 169-177.

Kodric-Brown, A., Sibly, R.M., & Brown, J.H. (2006). The allometry of ornaments and weapons. *Proceedings of the National Academy of Sciences of the United States of America*, 103, 8733-8738.

Macaranas, J.M., Colgan, D.J., Major, R.E., Cassis, G., & Gray, M.R. (2001). Species discrimination and population differentiation in ants using microsatellites. *Biochemical Systematics and Ecology*, 29, 125-136.

McArdle, B.H. (1988). The structural relationship: regression in biology. *Canadian Journal of Zoology*, 66, 2329-2339.

Mirth, C.K. & Riddiford, L.M. (2007). Size assessment and growth control: How adult size is determined in insects. *Bioessays*, 29, 344-355.

Morata, G. & Lawrence, P.A. (1979). Development of the eye-antenna imaginal disc of *Drosophila*. *Developmental Biology*, 70, 355-371.

Moreau, C.S. (2009). Inferring ant evolution in the age of molecular data (Hymenoptera: Formicidae). *Myrmecological News*, 12, 201–210.

Mosiman, J. & James, F. (1979). New statistical methods for allometry with application to Florida red-winged blackbirds. *Evolution*, 33, 444-459.

Nicholls, E., Rossi, M., & Niven, J.E. (2021). Larval nutrition impacts survival to adulthood, body size and the allometric scaling of metabolic rate in adult honeybees. *Journal of Experimental Biology*, 224(14), jeb242393.

Nijhout, H.F. & Wheeler, D.E. (1996). Growth models of complex allometries in holometabolous insects. *The American Naturalist*, 148, 40-56.

Nijhout, H.F., Riddiford, L.M., Mirth, C., Shingleton, A.W., Suzuki, Y., & Callier, V. (2014). The developmental control of size in insects. *WIREs Journal of Developmental Biology*, 3, 113-134.

Perl, C.D. & Niven, J.E. (2016). Colony-level differences in the scaling rules governing wood ant compound eye structure. *Scientific Reports*, 6, 24204.

Perl, C.D., Rossoni, S., & Niven, J.E. (2017). Conservative whole-organ scaling contrasts with highly labile suborgan scaling differences among compound eyes of closely related *Formica* ants. *Ecology and Evolution*, 7, 1663-1673.

Pusch, K., Seifert, B., Foitzik, S., & Heinze, J. (2006). Distribution and genetic divergence of two parapatric sibling ant species in Central Europe. *Biological Journal of the Linnean Society*, 88, 223-234.

Schlick-Steiner, B.C., Steiner, F.M., Konrad, H., Marko, B., Csosz, S., Heller, G., Ferencz, B., Sipos, B., Christian, E., & Stauffer, C. (2006a). More than one species of *Messor* harvester ants (Hymenoptera: Formicidae) in Central Europe. *European Journal of Entomology*, 103, 469-476.

Schlick-Steiner, B.C., Steiner, F.M., Moder, K., Seifert, B., Sanetra, M., Dyreson, E., Stauffer, C., & Christian, E. (2006b). A multidisciplinary approach reveals cryptic diversity in Western Palearctic *Tetramorium* ants (Hymenoptera: Formicidae). *Molecular Phylogenetics and Evolution*, 40, 259-273.

Schwarz, S., Narendra, A., & Zeil, J. (2011). The properties of the visual system in the Australian desert ant *Melophorus bagoti*. *Arthropod Structure and Development*, 40, 128-134.

Seifert, B. (2016). The supercolonial European wood ant *Formica paralugubris* SEIFERT, 1996 (Hymenoptera: Formicidae) introduced to Canada and its predicted role in Nearctic forests. *Myrmecological News*, 22, 11-20.

Shingleton, A.W., Das, J., Vinicius, L., & Stern, D.L. (2005). The temporal requirements for insulin signaling during development in *Drosophila*. *PLoS Biology*, 3, e289

Shingleton, A.W., Estep, C.M., Driscoll, M.V., & Dworkin, I. (2009). Many ways to be small: different environmental regulators of size generate distinct scaling relationships in *Drosophila melanogaster*. *Proceedings of the Royal Society of London. Series B*, 276, 2625-2633.

Smith, A., Fisher, B.L., & Hebert, P.D.N. (2005). DNA barcoding for effective biodiversity assessment of a hyperdiverse arthropod group: the ants of Madagascar. *Philosophical Transactions of the Royal Society B London*, 360, 1825-1834.

Sorvari, J. & Hakkarainen, H. (2009). Forest clear-cutting causes small workers in the polydomous wood ant *Formica aquilonia*. *Annales Zoologici Fennici*, 46, 409-416.

Spaethe, J. & Chittka, L. (2003). Interindividual variation of eye optics and single object resolution in bumblebees. *Journal of Experimental Biology*, 206, 3447-3453.

Steiner, F.M., Schlick-Steiner, B.C., Sanetra, M., Ljubomirov, T., Antonova, V., Christian, E., & Stauffer, C. (2005). Towards DNA-aided biogeography: an example from *Tetramorium* ants (Hymenoptera, Formicidae). *Annales Zoologici Fennici*, 42, 23-35.

Steiner, F.M., Schlick-Steiner, B.C., Konard, H., Moder, K., Christian, E., Seifert, B., Crozier, R.H., Stauffer, C., & Buschinger, A. (2006). No sympatric speciation here: multiple data sources show that the ant *Myrmica microrubra* is not a separate species but an alternate reproductive morph of *Myrmica rubra*. *Journal of Evolutionary Biology*, 19, 777-787.

Stevenson, R.D., Hill, M.F., & Bryant, P.J. (1995). Organ and cell allometry in Hawaiian *Drosophila*: How to make a big fly. *Proceedings of the Royal Society of London. Series B*, 259, 105-110.

Stockan, J.A., Robinson, E.J., Trager, J.C., Yao, I., & Seifert, B. (2016). Introducing wood ants: evolution, phylogeny, identification and distribution. In J.A., Stockan, and E.J.H., Robinson, (Eds), *Wood ant ecology and conservation* (pp 1-36). Cambridge University Press.

Thomas, R.H. (1993). Ecology of body size in *Drosophila buzzatii*: Untangling the effects of temperature and nutrition. *Ecological Entomology*, 18, 84-90.

Tschinkel, W.R. (2013). The morphometry of *Solenopsis* fire ants. *Plos One*, 8(11), e79559.

Vogler, A.P. & Monaghan, M.T. (2007). Recent advances in DNA taxonomy. J*ournal of Zoological Systematics and Evolutionary Research*, 45(1), 1-10.

Véle, A. & Modlinger, R. (2019). Body size of wood ant workers affects their work division. *Sociobiology*, 66(4), 614-618.

Wheeler, D.A. (1991). The developmental basis of worker caste polymorphism in ants. *The American Naturalist*, 138(5), 1218-1238.

Zagas, T. (1990). *Conditions for natural establishment of Pinus sylvestris in an area of Rhodopi Mountain, northern Greece* (In Greek). Ph.D. thesis, Aristotle University, Thessaloniki.

Zar, J.H. (1999). *Biostatistical analysis*. Prentiss-Hall, Inc. Upper Saddle River, N.J.

Received: February 03, 2021　　*Accepted: September 06, 2021*