

Article

Variability in the cephalothorax shape within and between populations of the spiny orb weaver spider *Gasteracantha kuhli*

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Abstract

Spiders are dominant sit-and-wait predators, and they are of potential value in biological control by feeding on other insects. The sizes and shapes of the spider's body are considered essential for their survival, especially in prey capture. The cephalothorax of the genus *Gasteracantha*, for example, is considered not only as a hierarchical status of their predation success but also as a defense mechanism from environmental disturbances. It is believed, therefore, that investigating how populations of a species vary, especially those collected from various geographical locations, is important. Thus, in this study, morphometric variations in the cephalothorax of *Gasteracantha kuhli* from selected local populations were assessed using relative warp analysis (RWA). Thirty-one landmarks were identified in the cephalothorax; coordinates were taken, Procrustes-transformed, and relative warp analysis was conducted. Results of RWA showed significant population variability in the cephalothorax shapes within and between *G. kuhli* populations, especially on the spines. The observed morphological differences can be argued to reflect their success on how well they do in their growth, survival, and reproduction in different environments where the spiders were found to dwell and are reflected in their external phenotype.

Keywords biological indicator; environmental disturbances; morphometric techniques; predators; variations.

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1 Introduction

Ecosystem dynamics can be influenced by local adaptation (Matthews et al., 2011), and argued to be reflected in morphological variations. It is for this reason that we conducted this study on the black-and-white spiny spider *Gasteracantha kuhli*, a species found inhabiting open forests and shrubby areas in selected Asian countries (Barrion and Litsinger, 1995; Koh, 2000; Kim and Park, 2015; Saitō, 1939; Tanikawa, 2007, 2009; Tikader, 1982; Simon, 1886-1904; Sebastian and Peter, 2009; Sheriffs, 1934). This species of spiny orb-

weaver spider build webs and stay at the center to wait for the prey (Platnick, 2010; World Spider Catalogue, 2019; Kim and Lee, 2012; Gupta et al., 2015). How populations of this species vary, especially those in various ecological habitats are wanting to be examined since much detailed morphological analysis for similar-looking species might reveal cryptic differences in selection pressures in natural environments. Understanding the possible role of the environment on the organisms' morphological diversity is a good starting point in understanding local adaptations and differentiation among species (Siepielski, 2010). Studies have shown that inter- and intraspecific variations in organisms can have significant effects on species' population, community, and ecosystem dynamics (Bolnick et al., 2011; Hausch et al., 2013; Vermeij, 1978). Thus, this study was conducted specifically in populations of the spiny-backed spider *G. kuhli* where they were found to be abundant in many different locations in Mindanao, The Philippines. Variability in morphological traits such as body size and shape provide us information and insights into how ecologically important species are in their natural habitat. Morphological differences can be argued to reflect their success on how well they do in their growth, survival, and reproduction in different environments as well as their roles in that environment populations (Bolnick et al., 2011; Hausch et al., 2013; Vermeij, 1978). It is for these reasons that we investigated population variability in *G. kuhli* by quantifying and qualitatively describemorphometric variations using geometric morphometric methods (GM), specifically the use of relative warp analysis. This technique that is very suitable in describing shape variation within and between many species quantitatively (Webster and Sheets, 2017; Gao et al., 2017; Presilda et al., 2018; Gualberto and Demayo, 2018; Sepe et al., 2019; Cabuga et al., 207a,b; Moneva et al., 2012a,b,c; Requieron et al., 2011; Sobrepena and Demayo, 2014). One advantage of using this method of analysis is that we can analyze more specimens and characterize shapes with higher fidelity making it an excellent way to quantitatively describe inter- and intraspecific variations in the abdominal shapes within, between, and among populations of *G. kuhli*.

2 Materials and Methods

G. kuhli was collected through opportunistic sampling from six locations in Mindanao, the Philippines. These are Tinago, Tomas Cabili, and Tominobo in Iligan City, Lanao del Norte; Balangao, Diplahan, Zamboanga Sibugay; Tigbao, Zamboanga del Sur and Dipolog, Zamboanga del Norte. The spiders were identified using the guide from the book of Barrion and Litsinger (1995). Images were captured using a Canon DSLR capturing the cephalothorax for image processing and subjected to geometric morphometric techniques.

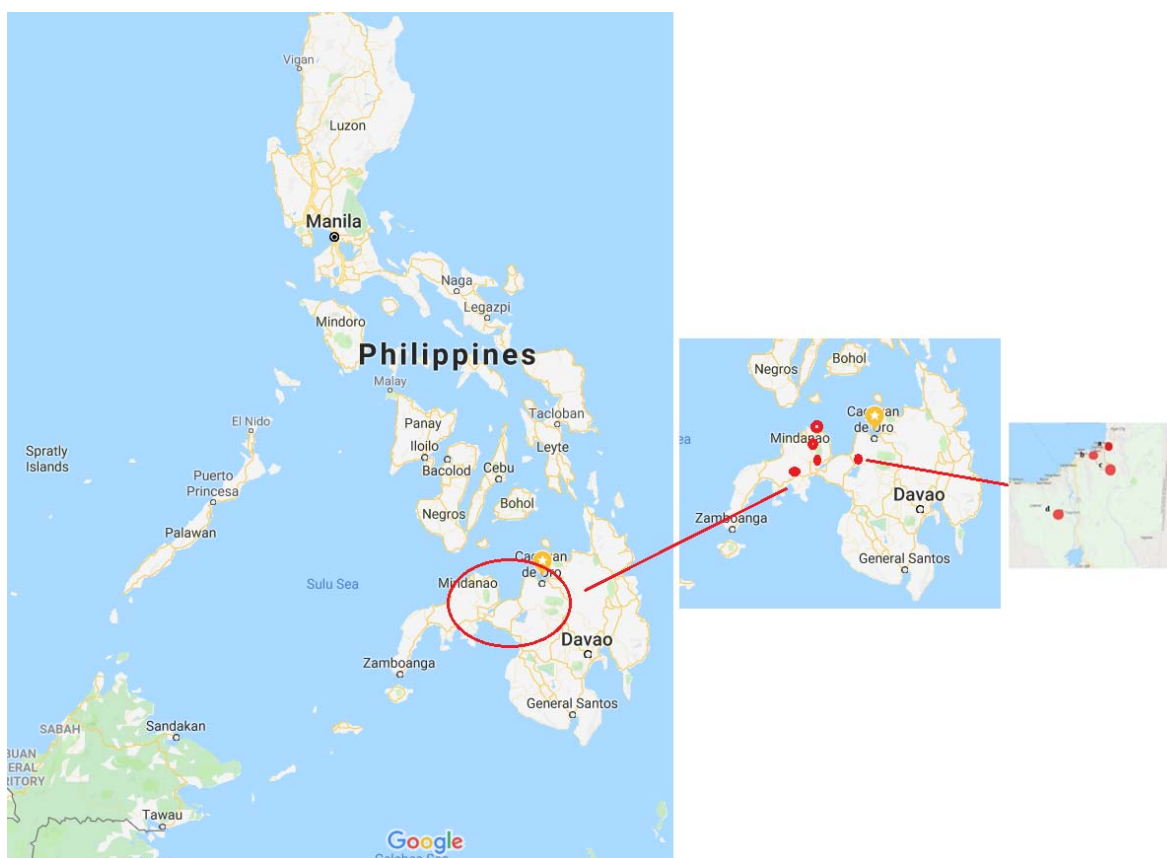


Fig. 1 Sampling area. A. Iligan City, Lanao del Norte (a. Tominobo; b. Baraas; c. Tomas Cabili; d. Tinago); B. Dipolog, Zamboanga del Norte; C. Tigbao, Zamboanga del Sur; D. Balangao, Diplahan, Zamboanga Sibugay.

There were a total of 31 landmarks along the border of the dorsal cephalothorax of the species forming an outline of the shape of the cephalothorax (Fig. 2, Table 1). These landmarks consist of the Procrustes shape coordinates of each specimen acquired through the use of the Tps Dig freeware 2.12 (Rohlf, 2008a). These landmark coordinates were then transferred to Microsoft Excel application for the organization of the data. These two-dimensional data was computed using the generalized Procrustes analysis (GPA) for the generalized orthogonal least squares using the tpsRelw ver. 1.46 software (Rohlf, 2008b), after which the principal components of the covariance matrix represented by the relative warps (RW) were computed using the alignment-scaling method that focuses on the unit centroid size. Representation was in the form histogram, and box plots are then generated from the acquired relative warp scores using the Paleontological Statistics (PAST) software version (Hammer et al., 2009). The Kruskal-Wallis test was used to analyze abdominal shape variations in populations of the two species. Canonical variance analysis (CVA) was also used to compare the patterns of variations among populations.

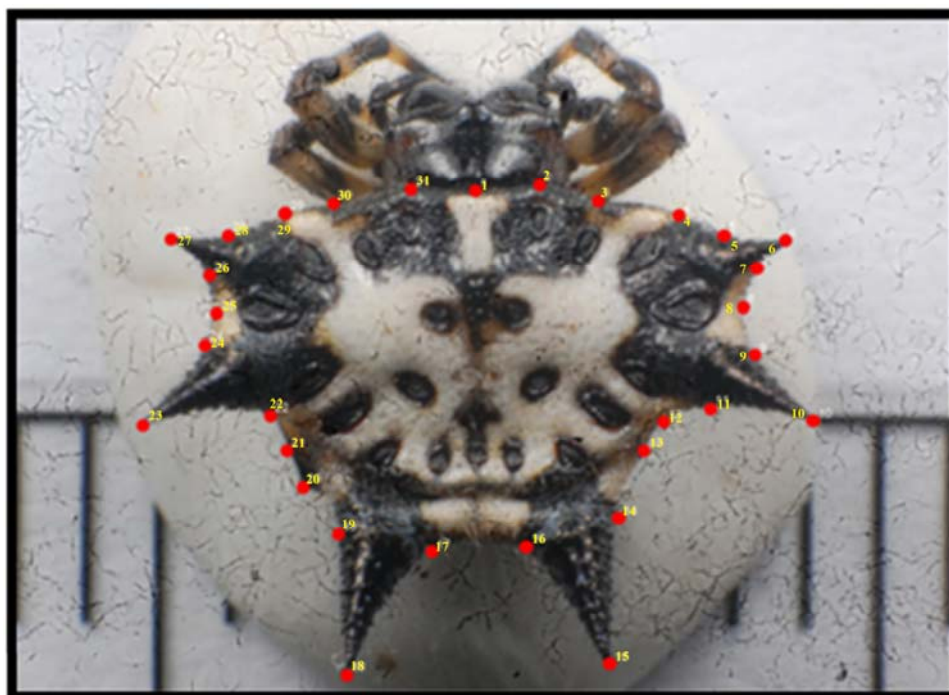


Fig. 2 Landmark points for the description of the cephalothorax shape of *G. kuhli*.

Table 1 Descriptions of the anatomical landmark (LM) points in the dorsal view of *G. kuhli* cephalothorax.

Landmark points	Description
1	The point on the midsagittal anterior margin
2-4	Curve points from the anterior midsagittal point to the base of the right anterior lateral spine
5	The base of the right anterior lateral spine
6	Tip of the right anterior spine
7	The base of the right anterior lateral spine
8	Curve point between the right anterior lateral spine and the left median lateral spine
9	The base of the right median lateral spine
10	Tip of the right median lateral spine
11	The base of the right median lateral spine
12	Curve points between the right median lateral and the right posterior-lateral spine
13	Curve point on the right posterior extension-like line of the cephalothorax carrying the posterior spines
14	The base of the right posterior-lateral spine
15	Tip of the right posterior lateral spine
16	The base of the right posterior-lateral spine
17	The base of the left posterior lateral spine
18	Tip of the left posterior lateral spine
19	The base of the left posterior lateral spine
20	Curve point on the left posterior extension-like line of the cephalothorax carrying the posterior spines
21	Curve points between the left median lateral and the left posterior lateral spine
22	The base of the left lateral median spine
23	Tip of the left lateral median spine
24	The base of the left lateral median spine

25	Curve between the left anterior and left median spines
26	The base of the left anterior lateral spine
27	Tip of the left anterior lateral spine
28	The base of the left anterior lateral spine
29-30	Curve points from the anterior midsagittal point to the base of the left anterior lateral spine
31	Curve on the flat anterior part of the cephalothorax near the anterior midsagittal point.

3 Results

Results of the CVA ($p=2.025E-308$) and Kruskal-Wallis test showed significant variations between populations of *G. kuhli* (Table 2). The CVA scatter plot (Fig. 3) shows the distribution of individuals in a population-based on the differences of the dorsal cephalothorax among the species of *G. kuhli*. It can be observed in the scatterplot that Balangao is differentiated from all other populations. Differences in the abdominal shapes of the spiders are shown in Fig. 4 and described in Table 3. It can be seen from the results that the differences observed in the abdominal shapes of the spiders can be quantitatively described using the landmark-based relative warp analysis of geometric morphometrics. What is interesting in the result is that populations that are geographically distant such as the Tominobo and Tigbao populations were clustered together, so is the Dipolog and Tomas Cabili populations. Likewise, Tomas Cabili and Tominobo populations that are not geographically distant were found to be different. Geographical distance therefore is not the sole basis for the variations between populations observed.

Table 2 Results of the Kruskal-Wallis test (p value) for significant differences in mean shapes of *G. kuhli*.

Relative warp	Population per species	Dipolog	Tigbao	Tomas Cabili	Tominobo
1	Balangao	1.87E-167*	1.72E-177*	2.10E-175*	3.62E-149*
	Dipolog		4.23E-58*	8.57E-19*	2.78E-17*
	Tigbao			8.09E-22*	1.65E-96*
	Tomas Cabili				4.79E-53*
2	Balangao	5.50E-162*	7.11E-177*	6.51E-178*	8.46E-173*
	Dipolog		1.50E-75*	1.64E-102*	8.76E-36*
	Tigbao			0.000155*	3.13E-16*
	Tomas Cabili				9.25E-31*
3	Balangao	4.89E-17*	2.70E-177*	5.37E-63*	6.32E-87*
	Dipolog		4.04E-176*	2.71E-28*	5.02E-53*
	Tigbao			2.70E-168*	1.23E-153*
	Tomas Cabili				4.18E-10*
4	Balangao	0.00011*	1.81E-177*	2.36E-63*	1.15E-68*
	Dipolog		9.81E-177*	8.70E-44*	3.05E-49*
	Tigbao			1.60E-166*	6.05E-160*
	Tomas Cabili				0.05755
5	Balangao	7.18E-172*	2.48E-177*	3.04E-161*	2.80E-96*
	Dipolog	0	1.16E-40*	6.19E-22*	3.01E-112*
	Tigbao	1.16E-39*	0	1.16E-82*	5.06E-152*
	Tomas Cabili	6.19E-21*	1.16E-81*	0	5.17E-71*

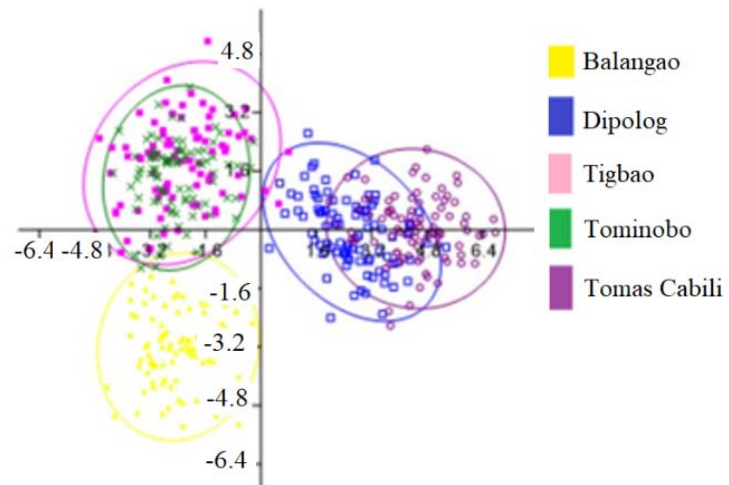


Fig. 3 CVA scatter plot of the dorsal abdominal shapes of five *G. kuhli* populations.

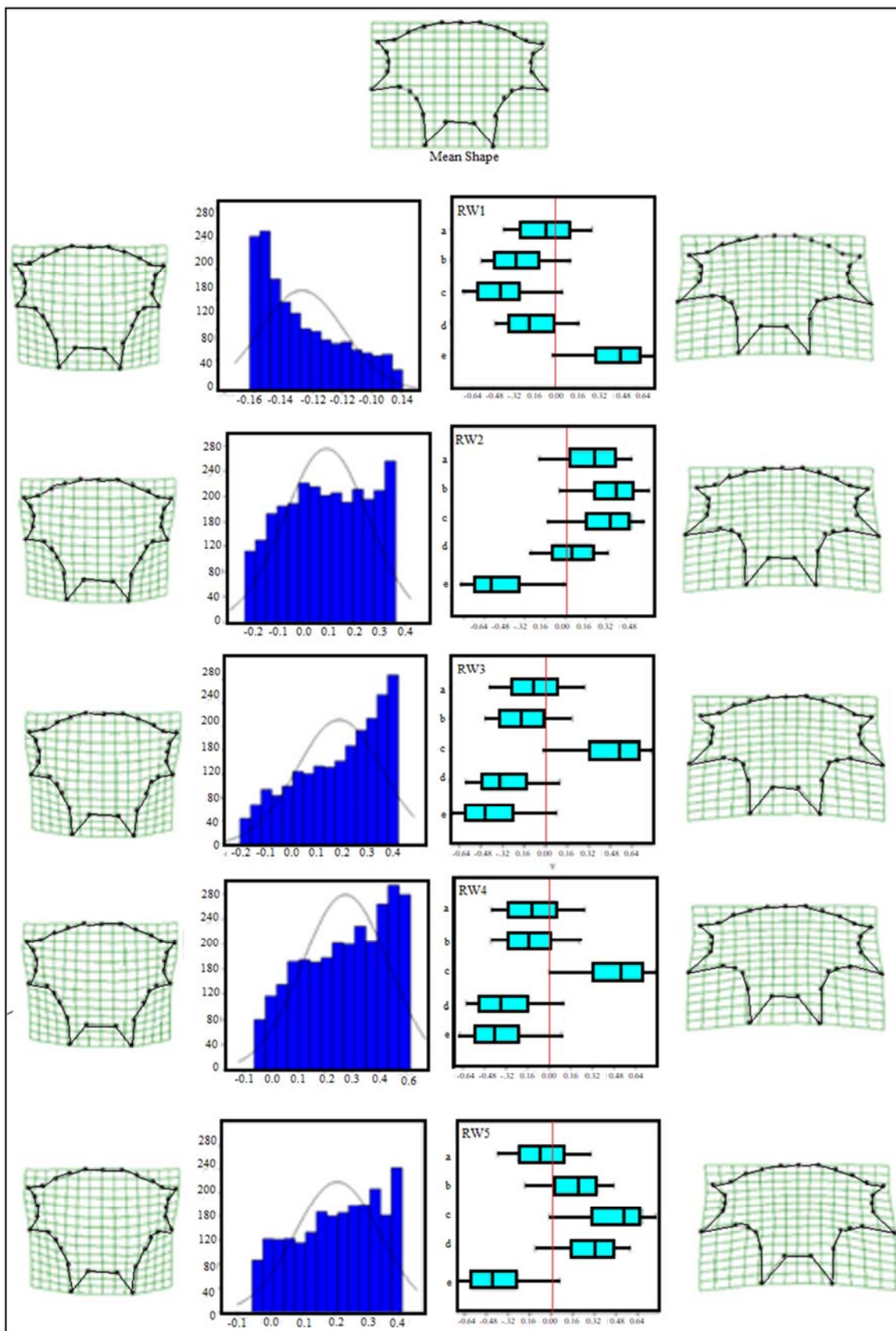


Fig. 4 Box plot and histogram showing variations in the shape of *G. kuhli*. a. Tominobo; b. Tomas Cabili; c. Tigbao; d. Dipolog; e. Balangao.

Table 3 Descriptions of the shapes of the dorsal cephalothorax of the five populations of *G. kuhli*.

RW	Variation	Dorsal cephalothorax
1	23.31%	Results of RW1 show the Balangao spiders have their cephalothorax's deformation on the posterior spines and more elongated vertically. The cephalothorax shows longer and larger spines in anterior, median, and posterior region. Transversely, the anterior part of the cephalothorax is wider and more convex in form. The overall formation of the dorsal cephalothorax is elongated horizontally. The other four spider populations have the outer the anterior outline of the body shape showing the convex shape and is shorter than the anterior outline, the curve distance between the lateral median spines has a lesser curve than the Balangao spiders. The anterior and the median spines are almost on the same size.
2	16.29%	The 2 nd relative warp scores show grid deformation in all of its spines. For the Dipolog <i>G. kuhli</i> spiders, the most prominent is the median spines, which got smaller, almost the same to the anterior spines resembling the mean shape. The space between the median and the posterior spines is also not that curved from the beginning of the left and right median spine going to the left and right and right posterior spines. For three other <i>G. kuhli</i> populations - Tominobo, Tomas Cabili, and Tigbao, all of the spines are larger, and transversely the anterior part of the cephalothorax is more elongated horizontally, with a more convex shape bending downwards, thus making the spines project downwards as well. The population from Balangao has spines that are smaller and transversely, the cephalothorax is wider when compared to the other three populations.
3	11.86%	The 3 rd relative warp shows the <i>G. kuhli</i> spider individuals from Tigbao has spines more elongated vertically. It is longer than wide. Variations can be observed in the anterior transverse part of the cephalothorax showing wider than long cephalothorax as well as longer and larger spines, the curve between the left and right median spines going down to the posterior left and right spines.
4	8.94%	The fourth relative warp shows four populations except for the spiders from Tigbao have more variance at the lateral side, especially in the anterior and median spines, which is visibly smaller from the mean and a longer posterior portion. The Tigbao spiders showed variances in all of the spines as well as showing larger and longer spines, transversely, the anterior part is wider than long, thus giving it an arched shape. The posterior end has narrower space.
5	6.05%	The 5 th relative warp showed the Balangao spiders vary from the other three other spider populations. The Tominodo was almost similar to the mean shape, while the Balangao spiders were showing a relatively small and thinner abdominal shape.

4 Discussion

The results that are shown in Figs 3 and 4, and Table 3, indicate the variations observable in abdominal shapes between populations using relative warp analysis. What is interesting in the results is that the variances in the abdominal shapes between populations cannot be directly attributed to geographical distance. It can be observed that geographically close populations were observed to be more different than those from geographically distant populations. This can be argued to be attributed to variations in many environmental factors such as predation, courtship, and defense (Bolnic, 2011; Hauschet al., 2013; Vermeij, 1978). Since the variations observed within, between and among the spiders were mainly on the size and shapes of the cephalothorax and spines, it can be argued that these variances in the direction and size of the cephalothorax and spines play a big role in their defense against predators (Fig. 3 and 4). Very conspicuous variations observed in the spines either smaller and shorter or as long and larger spines, maybe attributed to the

environment where these spiders were collected. The variations in shapes and sizes of the spines, as well as the cephalothorax shapes within and between populations, can be attributed to the spider's differences in adapting to their environment. While there are just a few documentation about the biology of *Gasteracantha* species, especially on their biology and behavior (Huma, 1971), the observations made on the species populations of having an eating habit of sit and wait at the center of the web while waiting for the prey to be trapped (Kemp et al., 2013) can be a basis for the variations observed in the sizes and shapes of the cephalothorax of the spiders. This species can eat more significant individuals of other species by wrapping the prey with silk (Huma, 1971; Muma and Stone, 1971) and since their cephalothorax serves as an essential part of their foraging success, the observed variations in size and shapes of the cephalothorax mainly the spines maybe a product of adaptation of the species. Aside from using eye-catching displays of color to attract prey (Vermeij, 1994) and the need to defend themselves against potential predators such as birds, bigger animals, and other environmental disturbances, their hard colorful cephalothorax, and their spines serves an essential role for their defense against these organisms. Their spines serve as an anti-predator function; thus, the observed variations in their spines within populations may reflect the extent of predatory protection by this species, which may vary from locality to locality and not on the scale of geographical distance where these spiders were collected.

5 Conclusion

Results of RWA showed significant population variability in the cephalothorax shapes of the *G. kuhli*, especially on the spines. Ecological/environmental variations were most likely contributed to the variances in size and shapes of the cephalothorax including the spines generated by relative warp analysis

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