OBSERVATIONS ON GEOGRAPHIC VARIATION IN THE ASIAN FROG, *HOPLOBATRACHUS RUGULOSUS* (ANURA: RANIDAE)

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(with three text-figures)

ABSTRACT.– **This study examines a series of morphometric, meristic, and colouration traits in the wide-ranging *Hoplobatrachus rugulosus*. Comparison of four samples (Myanmar, Thailand, Hong Kong, Taiwan) reveals minor differences within and among the sample localities, but the differences do not display concordant patterns of geographic variation of the twenty-three traits examined. All populations display sexual dimorphism in morphometric traits, no dimorphism in hindfoot webbing or dorsal rugosity, and dimorphism of colouration in Taiwan and Myanmar populations. Discriminant analysis of the morphometric traits provides evidence of modest segregation of the sample localities, but the small sample sizes make these results equivocal. Eastern *H. rugulosus* averages larger and more rugose than western populations. Taiwan frogs usually have spotted bellies, and spotting decreases westward to immaculate bellies in Myanmar frogs. Hong Kong frogs have less hindfoot webbing than the three other populations. Other traits differ, but their geographic patterns do not match that of the preceding ones or display another single pattern.**

KEYWORDS.– **Anura, Ranidae, *Hoplobatrachus rugulosus*, geographic variation, sexual dimorphism, morphometry, Hong Kong, Myanmar, Taiwan, Thailand.**

INTRODUCTION

Many species of Asian frogs have distributions that extend from the Indian subpeninsula to the coast and islands of eastern Asia. Some of these species clearly are composites, consisting of numerous similar appearing allopatric species, for example, the *Fejervarya limnocharis* complex (Dubois and Ohler, 2000). Other presumably widespread species are less obvious composites, and their composite nature is revealed only by close examination of behavioral, molecular, or morphological characteristics (e.g., *Kalophrynus interlineatus* and *K. pleurostigma*; Matsui et al., 1996). These two extremes of hidden (unrecognized) species suggest that many more of the widespread Asian frogs consist of multiple species.

Frog monitoring at the Chatthin Wildlife Sanctuary in north-central Myanmar (Zug et al., 1998) and the joint California Academy of Sciences – Smithsonian Institution and Myanmar Wildlife Division herpetofaunal survey have encouraged a systematic examination of the Myanmar anuran fauna, both because of the discovery of new species and the need to identify correctly the resident species. *Hoplobatrachus rugulosus* or the Chatthin frogs that fit the "*rugulosus*" paradigm are a major component of the Chatthin paddy frog community. Being large frogs with numerous dorsal longitudinal ridges, they are easily labeled *rugulosus*. But is this name designation correct? Specifically, are Myanmar "*rugulosus*" populations part of a genetic continuum from the nominal population in the Hong Kong area? The following analysis is a preliminary investigation of this question through an examination of regional variation in external morphology.

MATERIALS AND METHODS

We selected four geographic areas for analysis: Taiwan (TA); Hong Kong (HO; type locality of *Hoplobatrachus rugulosus*); Thailand (TH); and Myanmar (MY); see list of specimens examined in the Appendix I. These four localities encompass the longitudinal expanse of this species' distribution, although they certainly do not encompass all the variation therein. We limited our sample selection to the availability of series in the United States National Museum's collections (USNM), with the addition of a Hong Kong topotypic series from the Museum of Comparative Zoology, Harvard University (MCZ). These samples provide adequate adults for interpopulational comparisons. For our comparison, we specifically restricted our samples to adults from a single geographic area in order to limit intraregional variation. Additionally, all data were recorded by the first co-author to ensure consistency, and as recommended by Hayek et al. (2001), a single specimen (USNM 132075, adult female, Taiwan) was measured multiple times (15) to provide an estimate of intra-observer variation. This last data set was captured on five different days and at three different times during a session measuring frogs from other samples.

Our characters include sets of quantitative (mensural and meristic) and qualitative (colour pattern) features. The characters include the basic frog mensural set as well as additional measurements and traits that appeared on an initial examination to characterize "*rugulosus*." The characters are:

Mensural. [mm; all bilateral measurements recorded from the right side.] Snout-vent length (SVL): The distance from the tip of snout to the vent. The measurement is taken with the frog's venter adpressed against a flat, firm surface. Head length (HeadL): Straight-line, horizontal distance from the tip of snout to the posterior corner of the jaws. Head width-mid (HeadWm): Straight-line, transverse distance from the left to right edges of the lips at the posterior edge of the eyes. Head width-posterior (HeadWp): Straight-line, transverse distance from the left to right edges of the corner of the jaws. Internarial distance (Intnar): Distance between the left and right nares. Naris-eye distance (NarEye): Distance between the naris and the anterior corner of the orbit. Eye-ear distance (EyeEar): Distance between the posterior corner of the orbit and the anterior, internal border of tympanic annulus. Tympanum diameter (Tymp): Horizontal diameter of the tympanum from outer edge of annulus. Trunk length (TrnkL): Straight-line, horizontal distance from axilla to inguen. Trunk width (TrnkW): Transverse distance from outer edges of left and right sacral diapophyses. Forelimb length (ForlL): Straight-line distance from elbow to wrist. Thigh length (ThghL): Straight-line distance from vent to knee. Crus length (CrusL): Straight-line distance from knee to ankle. Tarsus length (TarsL): Straight-line distance from ankle joint to heel. Hindfoot length (HndfL): Straight-line distance from heel to tip of 4th toe.

Meristic. Hindfoot webbing: coding of Savage and Heyer (1997), with the exception that webbing is recorded only in halves and whole numbers. Toes are identified by Roman numerals, and inner (anterior) and outer (posterior) edges by integers; for example, the inside edge of third toes is III1, its outside edge III2. Glandular fold development; folds are glandular elevations 5X or more longer than wide. Anterior body folds (AntFold): number of folds across dorsum at level of forelimb insertions. Posterior body folds (PstFold): number of folds across dorsum at level of sacral diapophyses.

Qualitative Colour pattern. These traits are coded arbitrarily in a numerical sequence. Dorsum (DorsC), three states: unicolour [0]; spotted or blotched [1]; reticulate [2]. Thigh (ThigC), rear surface: four states: reticulate [0]; vertical bars [1]; spotted or blotched [2]; horizontal stripe(s) [3]. Chin & throat, midline (ChinMC): longitudinal stripe, present [1] or absent [0]. Chin & throat, lateral (ChinLC): immaculate [0]; lightly spotted [1]; moderate to strongly spotted or reticulate [2]. Chest (ChesC): three states: immaculate [0]; spotted or blotched [1]; reticulate

[2]. Belly (BelC): immaculate [0]; spotted or blotched [1]; reticulate [2]. All observations on colour pattern derive from preserved specimens.

We examined the gonads of all specimens to determine maturity and sex. We used SYSTAT version 9 for all statistical analyses; Student's *t*-test compared means of adult female and male mensural and meristic variables for identifying sexual dimorphism; ^{2} test provided tests of colour pattern sharing among samples; principal components analysis (PC) performed with a correlation matrix and no rotation; and linear discriminant function analysis (DA) with a step-wise model-backward entry of variables provided evaluations of regional differentiation. Unless noted otherwise, all analyses used un-transformed data.

RESULTS

Mensural.- The intra-observer variation was modest. The adult female selected for repetitive measurement ($n = 15$) is one of the two largest females in our samples. The coefficient of variations (*CV*) for her variables range from 0.008 to 0.091: *CV* 3% SVL, TrnkL, HeadL, HeadWp, Intnar, NarEye, CrusL, TarsL, HndfL; *CV* = 4-6% TrnkW, HeadWm, EyeEar, Tymp, ThghL; *CV* = 9 ForlL. SVL has the lowest *CV* (0.8%) and a standard deviation (*s*) of 0.85 mm, indicating an accuracy of 1 mm for this measurement. The *s* of TrnkL, TrnkW, HeadL, HeadWm, ForlL, and ThghL exceed 1.0 mm (1.2-1.8). These *CV* are less than those of the individual sample localities. For example, the larger (n 9) male (TA, TH, MY) and female (TA, MY) samples have CV ranges of 0.054-0.186, 0.070-0.161, 0.102-0.130, 0.085-0.190, 0.106-0.191, respectively. With the exception of Taiwan male sample, *CV*s are predominantly 10%, although 16%; the Taiwan males are mostly < 10%. The *CV*s show males to be less variable than females, that is, they have a narrower size range as adults.

A comparison of the adult females and males in each sample (Table 1) reveals that the males average smaller than females at all localities, although the means between the sexes are significantly different (pooled variance $p < 0.05$) only for the Taiwan and Myanmar samples. This re-

sult highlights the role and necessity of nearly equal sample sizes for statistical confirmation of sexual dimorphism. The result further demands that all subsequent interregional comparisons be among individuals of the same sex.

PC analysis provides an independent assessment of adult dimorphism (Fig. 1) at each locality and the possibility of identifying variables with the greatest discriminatory potential. All four localities have the first component (PC1) denoting size. Although there is overlap between females and males, males typically are negative and females positive on the PC1 axis (Fig. 1 left). Females and males overlap strongly on PC2. All characters load heavily (0.44-0.99) on PC1 at all localities, and most loadings are > 0.80 (Table 2). Size (PC1) accounts for 72.4%, 80.3%, 76.7%, and 90.2%, respectively (Taiwan, Hong Kong, Thailand, Myanmar), of the total variance of the measurements. Either CrusL or SVL are the highest loading variables for PC1 in the four samples, but often, other variables are nearly equal in this component loading. PC2 loadings are mostly low (-0.30 to 0.30 for 75% or more of the variables in each sample), and account for 6.9%, 9.2%, 7.8%, and 2.3% of the variance, respectively. The variables with the highest loading on PC2 are Tymp and Intnar (Taiwan), ThghL and TrnkL (Hong Kong), ThghL (Thailand), and ThghL (Myanmar). Log transformation of the data did not alter the amount of segregation among the sexes; it simply reversed the signs of the components. On this basis, transformation was not used in subsequent analyses.

PC comparison of adult females and males (Fig. 1 right; results presented for males only) independently from all four localities show no discrete clustering for any locality. Again, all variables load heavily (0.75-0.98) on PC1 and much less so on PC2 (Table 2). The highest loading PC2 variables for females (see Table 2 for males) are EyeEar (0.50), TrnkL (-0.48), and ThghL (-0.46); all other variable loadings are 0.26 to 0.002. PC1 accounts for 77.8% and PC2 for 5.9% of the variance in females, and 67.7% and 8.4%, respectively, in males.

DF analyses of the samples examined the differentiation of the four localities using all vari-
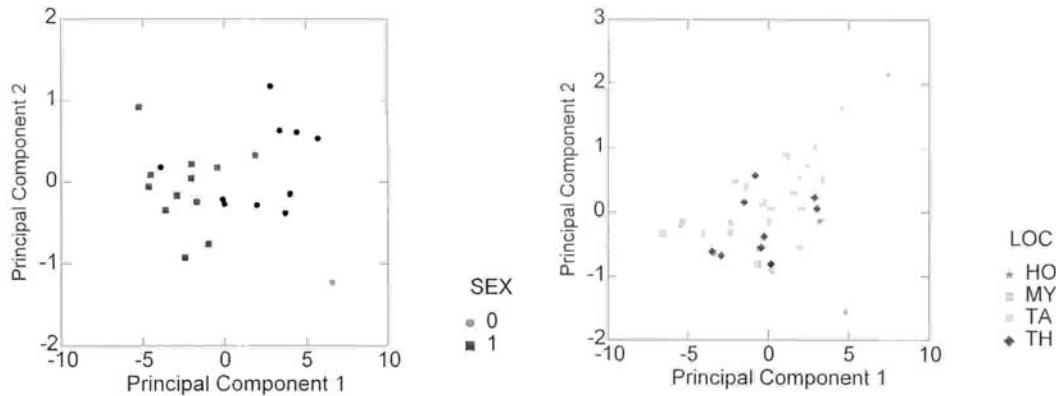
FIGURE 1: Principal component analyses of *Hoplobatrachus rugulosus*. Left. A comparison on adult females and males from Myanmar; females represented by circles, males by squares. Right. A comparison of adult males from the four Asian localities: Taiwan, squares; Hong Kong, stars; Thailand, diamonds; Myanmar, circles.
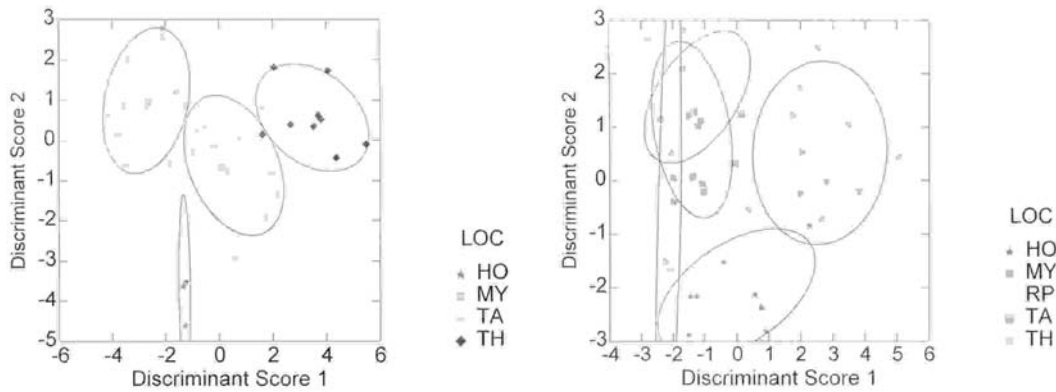

FIGURE 2: Discriminant function analyses of adult male *Hoplobatrachus rugulosus*. The variables used in the final stepwise DF model are in Table 3; the confidence ellipse defines $p = 0.68$ of the sample. The clusters are: Taiwan, squares, open squares for repeat-measured female; Hong Kong, stars; Thailand, diamonds; Myanmar, circles.

ables except ForlL and a subset consisting of the highest loading variables in the PC analyses, i.e., SVL, CrusL, TrnkL, ThghL, Tymp, and Intnar. Stepwise DF of the "total" variable set identified five variables for females and four for males (Table 3) as the "best" ones for the discriminatory models. Only the HeadWp and ThghL variables are shared between the two sexes. These reduced variable sets yield an overall classification of 89% (83% jackknifed) for females and 94% (83%) for males (Table 3). The sample-locality clusters observed on bivariate plots of the first three DF scores for females show modest segregation of Taiwan, Hong Kong, and Myanmar (Fig. 2 left); the Thai sample consists of only

three individuals and overlaps the Taiwan cluster. Males also show modest segregation (Fig. 2 right) of the four localities, also with slight cluster overlap. For males, the smallest sample (Hong Kong) is distinctly separated from the other three localities. Using the six PC-identified variables, only the Thailand sample yields 100% correct classifications (standard & jackknifed) in the female analysis, and 100% and 89%, respectively, in the male analysis; the average overall jackknifed accuracy is low (69, 67%) for both females and males. Adding the repeat-female sample to the female DF stepwise model increases the size (variation) of the Taiwan cluster (Fig. 2 left), but the repeat measures do not overlap with

TABLE 1: Select measurements of the four samples of *Hoplobatrachus rugulosus*. Abbreviations and definition of characters are explained in the Materials and Methods section. * denotes significantly different means for female and male samples, using a pooled variance $p < 0.05$.

	N	SVL	Head Width-mid	Tympanum Diameter	Crus Length
Taiwan					
female	14	95.9 ± 9.18*	26.6 ± 2.61*	6.7 ± 0.57*	39.9 ± 4.11*
male	12	81.1 ± 4.34	22.9 ± 1.53	6.2 ± 0.54	35.2 ± 2.34
Hong Kong					
female	8	105.2 ± 14.31	29.5 ± 5.10	7.2 ± 1.15	45.2 ± 7.11
male	3	90.4 ± 8.02	25.9 ± 1.15	6.5 ± 0.58	41.3 ± 2.85
Thailand					
female	3	84.4 ± 7.88	25.4 ± 3.87	5.8 ± 0.55	38.6 ± 5.30
male	9	77.2 ± 5.83	22.6 ± 2.32	5.5 ± 0.50	35.2 ± 3.59
Myanmar					
female	11	88.3 ± 11.07*	23.8 ± 2.53*	6.9 ± 0.98*	38.1 ± 4.50*
male	12	71.3 ± 7.4	18.8 ± 2.04	5.3 ± 0.66	30.4 ± 3.14

TABLE 2: Principal component loading values for the comparisons of morphometric characteristics of Myanmar adult females and males (Fig. 1 top) and of adult males of *Hoplobatrachus rugulosus* for the four sample locality sites (Fig. 1, bottom). The loading values are similar for the female-male comparisons at the other sites and for comparison of females among the four sites.

	Females & Males		Sample Localities	
	PC1	PC2	PC3	PC4
SVL	0.99	0.06	0.97	0.03
HeadL	0.95	0.10	0.85	0.07
HeadWm	0.96	0.17	0.90	-0.27
HeadWp	0.99	0.12	0.90	-0.33
Intnar	0.92	-0.25	0.81	0.08
NarEye	0.93	0.12	0.76	0.31
EyeEar	0.91	0.08	0.67	-0.33
Tymp	0.92	-0.23	0.74	0.25
TrnkL	0.94	0.05	0.57	0.61
TrnkW	0.96	0.02	0.92	-0.15
ForlL	-	-	0.81	0.39
ThghL	0.90	-0.34	0.58	0.35
CrusL	0.98	-0.001	0.95	-0.16
TarsL	0.98	0.02	0.87	-0.18
HndfL	0.98	0.04	0.90	-0.24

TABLE 3: Summary of linear discriminant function results for interpopulational comparison of *Hoplobatrachus rugulosus*. Females and males were compared separately owing to sexual dimorphism of mensural traits. Abbreviation: J%, jackknifed classification.

	Males			Females		
Variables in final model						
	HeadWp			HeadL		
	NarEye			HeadWp		
	ThghL			EyeEar		
	HndfL			ThghL		
				CrusL		
Classification accuracy						
Locality	n	%	J%	n	%	J%
Taiwan	12	92	67	14	93	93
Hong Kong	3	100	100	8	63	50
Thailand	9	89	89	3	100	100
Myanmar	12	100	92	11	100	91
Total	36	94	83	36	89	83

Myanmar and Hong Kong. The repeats, however, indicate that larger samples of each of these three localities would increase the overlap among them.

Meristic.- Webbing of the hindfoot shows no sexual dimorphism within samples; the probabilities for all *t*-tests exceed 0.08 and most are > 0.30. The extent of interdigital webbing varies between localities (Table 4). Hong Kong adults have less webbing than adults of the other localities, and although variation in webbing occurs

TABLE 4: Variation in extent of hindfoot webbing in the four samples of *Hoplobatrachus rugulosus*. The data are means for each sample; the abbreviations are explained in the Materials and Method section.

	n	I	II1	II2	III1	III2	IV1	IV2	V
TA	30	0.78	2.00	0.87	2.20	1.57	2.97	3.33	1.77
HO	18	0.67	2.00	0.53	2.17	1.14	2.83	3.00	1.00
TH	18	0.97	2.00	0.84	2.38	1.36	2.94	3.22	1.28
MY	25	0.82	2.00	1.00	2.47	1.56	3.00	3.47	1.72
Greatest Difference		0.30	0.00	0.47	0.20	0.43	0.17	0.47	0.77

TABLE 5: The density of glandular body folds on the trunk of adult *Hoplobatrachus rugulosus*. The mean number of body folds is presented on the mid-diagonal of the locality-locality matrix; first number is PstFold, the second AntFold. The right upper half of the matrix shows significant difference between AntFold of paired localities, and lower left half the significance difference between PstFold. $* = p = 0.05\text{-}0.002$, $** = p \leq 0.001$, $- = p > 0.05$.

	n	TA	HO	TH	MY
TA	26	9.7\10.0	*	-	**
HO	11	*	6.5\7.6	-	-
TH	12	*	-	8.1\8.7	*
MY	23	**	-	**	6.0\6.9

among these latter localities, they are more similar to one another than each is to Hong Kong. The greatest difference in web attachment among the localities is for the fifth toe (V), roughly a difference of three-quarters of a phalanx in attachment. Although hindfoot webbing in Hong Kong frogs is reduced relative to the other populations, webbing is still well developed.

We quantified rugosity by counting the number of glandular folds at the shoulder (AntFold) and the "hips" (PstFold). As for toe webbing, there is no evidence of sexual dimorphism but rugosity has high intrasample variation (AntFold $CV = 19\text{-}41\%$, PstFold $CV = 19\text{-}63\%$). In all samples, the glandular folds average more numerous anteriorly than posteriorly (Table 5). Our westernmost and easternmost samples, respectively, are the least and most rugose. Samples from Taiwan are significantly more anteriorly rugose than those from Hong Kong and Myanmar, and posteriorly more rugose than the other three samples. Even though samples from Hong Kong and Thailand are not significantly different from one another, the rugosity of those from Hong Kong is more similar to those from Myanmar than Myanmar is to its nearest neighbour Thailand.

Colour.- Our colouration variables document overall dorsal colouration (DorsC), thigh pattern (ThghC), and ventral colouration in four areas (ChinMC, ChinLC, ChesC, BelC). Because we arbitrarily assigned numerical values to different patterns (e.g., spots vs. immaculate) within each colour variable, we can test the similarity of patterns between sexes and among localities by χ^2 analysis of frequency tables. There is only slight evidence for sexual dimorphism in colouration and that is in a single trait, DorsC, and at one locality, Myanmar. All Myanmar males are spotted but only 64% of the females ($df\, 1$, $\chi^2 = 5.28$, $p = 0.02$). Only Taiwan also has adults without spots or blotches on the dorsum, although these dorsally unicoloured individuals are all females, the

TABLE 6: Summary of adult colouration of *Hoplobatrachus rugulosus* from Taiwan, Hong Kong, Thailand, and Myanmar. The abbreviation for variable names are in the Materials and Methods section; all values are percent of total adults at a locality. An asterisk denotes that the frequency distribution of colour states among localities is significantly different, $p < 0.01$.

	DorsC unicolour	spotted	ThghC* reticulate	spotted	ChinMC* absent	present	ChinLC light	moderate	ChesC* immaculate	spotted	BelC* immaculate	spotted
TA	12	85	36	66	20	80	20	80	0	100	24	76
HO	0	100	100	0	36	64	9	91	27	73	73	27
TH	0	92	58	33	0	100	0	100	0	100	83	17
MY	17	83	87	13	0	100	13	87	52	48	100	0

larger Taiwan sample does not yield significance (df 1, ^{2} = 2.95, p = 0.23).

Examining adult colouration among the samples (Table 6) shows that frequency distribution of the colour states for DorsC and ChinL are statistically equivalent among the four localities, and the distributions of states are different for the other four colour traits. There is no striking concordance of colour patterns among the localities. The pattern on the rear of the thigh is predominantly reticulate at Hong Kong, Thailand, and Myanmar. A distinct midline stripe occurs on the chin of all Thailand and Myanmar individuals, but on two-thirds of the Hong Kong and three-quarters of the Taiwan frogs. Most frogs in all populations have moderate to heavy markings on the sides of the chin. The chest of Taiwan, Hong Kong, and Thailand always or usually bear some spots or streaks, but only about half of the Myanmar frogs have markings on the chest. The belly of most Taiwan frogs is marked, and this marking becomes less frequent westward with the Myanmar frogs lacking any belly marks.

DISCUSSION

Mensural.- Hayek et al. (2001) recommended 20 repeated measures of the same individual to examine intra-observer variation. Our sample size is somewhat smaller (n = 15), but our results match their result and, thus, appear adequate. SVL has the lowest variation (CV < 1%), and similarly all our measurements, except ForlL, have $CV \leq 6\%$ as in their study (Hayek et al., 2001: Table 4). This low variation argues for measurement consistency within our data set and shows our ability to obtain fairly reliable data sets from specimens that are variably preserved and positioned. We also interpret the low variation of the repeated-measures data as confirmation that the higher variation observed within the locality samples reflects the natural variation of adult *Hoplobatrachus rugulosus* populations, although the repeat data remind us that larger samples will increase the range of variation and likely reduce the segregation of samples in multivariate space. Thus, the repeat data make us more cautious in the interpretation of a sample's

uniqueness, especially for the small sample sizes of this study. Because *CV* is sample size dependent, the variation of our samples is increased by their small size.

Size sexual dimorphism is a common aspect of anuran morphology (Zug et al., 2001). Our inability to confirm this dimorphism statistically in the Hong Kong and Thailand samples likely results from few individuals of one sex and the resulting strong inequality of sample size between the sexes. With the exception of Thailand sample, females average about 15 mm larger than males (Table 1), and this difference occurs in the populations with the largest (Hong Kong) and smallest (Myanmar) individuals. Our data suggest a somewhat clinal decrease in size from east to west.

Neither the PC nor the DF analyses of the morphometric variables argue strongly for regional differentiation of *H. rugulosus* populations. The PC results segregate individuals on the basis of size on the first component. The second component also appears to size influenced, with head size being most important for the Taiwan population, and trunk and thigh length for the other populations. The DF results also show an influence of head and hindlimb variables.

The modest segregation of localities through DF analyses (Fig. 2) hints at some regional differentiation. With our bias toward Myanmar *H. rugulosus*, we see the nonoverlap of Thailand and Myanmar samples as a suggestion of speciation of Myanmar *rugulosus*; however, until samples are enlarged and their number increased, any nomenclatural change is inappropriate. Similarly, the differences of the Hong Kong sample require a detailed comparison with mainland China populations because Hong Kong is the nominate population of *H. rugulosus*.

Meristic.- Hindfoot webbing shows no sexual differentiation but shows regional differences (Table 4). Hong Kong frogs have the least amount of webbing, but this least is relative because webbing is still extensive on the hindfeet. Webbing is usually recorded in smaller fractional units (Savage and Heyer, 1997) than halves;

however, preliminary data collection showed our accuracy (consistency) was higher at half-unit recording. We are unable to judge the influence of this aspect of data-capture on observed regional difference.

Rugosity also lacks evidence of sexual dimorphism and shows evidence of regional differentiation (Table 5) with Taiwan frogs being the most rugose and Myanmar ones the least. This most to least rugosity is not, however, a cline. Hong Kong and Thailand frogs are intermediate but Hong Kong rugosity is more similar to the Myanmar condition than it is to Thailand's.

Colour.- Of the six colouration variables, only dorsal colouration hints at sexual dimorphism and only in the Taiwan and Myanmar samples. In both, some females have unicolour or near unicolour backs. None of the other colouration variables show sexual dimorphism, and there is no strong signal of regional differentiation in colouration although some regional differences exist (Table 6). The rear of the thigh in Taiwan frogs is usually spotted, whereas it mostly reticulate in Hong Kong and Myanmar frogs, somewhat less so in the Thailand sample. Taiwan and Hong Kong frogs occasionally (20 and 36%) lack the midline chin stripe, which is invariably present in the Thailand and Myanmar individuals. Only belly colouration displays a geographic trend with the sequential loss of spotting on the belly from east to west.

Conclusions.- Our original question, "Are Myanmar "*rugulosus*" populations part of a genetic continuum from the nominal population in the Hong Kong area?," cannot be answered unequivocally by our analysis. The analyses of regional differentiation within the four sets of morphological characters reveal differences between each of the four populations examined. These differences, however, do not support a hypothesis of unquestionable regional differentiation and can as easily be interpreted as a genetic continuum as speciation. Mensural data shows sexual dimorphism if the samples are sufficient large. Additionally, eastern populations average larger than the western ones (Thailand, Myanmar). Webbing suggests differentiation of Hong Kong *H. rugulosus* from the other localities. Rugosity and colouration show differences among the samples and hint at regional differentiation. The small sample sizes likely contributed to our inability to discern regional differentiation. We predict that the patterns seen within and between samples will be re-enforced by larger samples. We also predict that the addition of new localities will introduce new patterns of intrasample variation. Our data do not declare the absence of regional genetic differentiation in *Hoplobatrachus rugulosus*, rather they indicate that such differences cannot be verified by external morphology alone. Behavioral and molecular data also are required.

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APPENDIX I

Examined specimens.- Arranged from west to east. Taiwan: USNM 38209-211, 38218-219, 132075, 133157, 139958-962, 141635-651. Hong Kong: MCZ 109353-359, 115680-682, 115685, 115688-690, 115692, 117035, 117037; USNM 37512. Thailand: USNM 206086, 206090-091, 206093-094, 206096-106. Myanmar: USNM 520470-489, 524039-042, 537465.