

SIZE VARIATION IN BUFO MELANOSTICTUS FROM JAVA AND BALI (AMPHIBIA)

by

GILBERT CHURCH

Department of Biological Sciences
Medical Faculty University of Indonesia, Djakarta

Introduction

Although *Bufo melanostictus* SCHNEIDER is distributed widely and abundantly from India and the Himalayas to Southern China, and south into the Malay Peninsula and Archipelago (BOULENGER, 1912; VAN KAMPEN, 1923), no attempt at analysis of local populations of this species has been undertaken. Its neglect can almost be measured by its prevalence, as though its ubiquity made it unimportant in the eternal search for new species in new geographical areas. Among later writers, for example, SMITH (1930) has nothing to add to the description of the species given by BOULENGER (1912); and LIU (1950) might just as well have omitted it entirely from his study. Yet because of its commonness, its ability to invade and succeed in almost all habitats, and the ease with which it can be collected, *melanostictus* offers an opportunity for a study of variation within populations of a single species from different localities which is unique among the Amphibia.

This report presents the results of analyses of snout-vent measurements of 3,908 male and female *B. melanostictus* from sixteen different local populations on the islands of Java and Bali. Fifteen series were collected in localities ranging from the western to the eastern extremities of Java; and one series was collected in West Bali in an area to which the species has been found by CHURCH (1959) to be limited. It is hoped that an opportunity to extend the study to Sumatra, Borneo and the Malay Peninsula will present itself in the future.

The writer wishes to acknowledge the help in making these collections of his two research assistants, SIE KIAN TJONG and TJIOE DJIN YONG; the kindness of His Highness TJOKORDA AGUNG GDE SUKAWATI of Ubud and Dr. J. N. POST of Negara, Bali; and Dr. D. A. TISNA AMIDJAJA and Dr. CHRISTOPH ZELLER of Bandung. The enthusiasm and optimism of Mr. A. M. R. WEGNER, Zoological Museum, Bogor, is always a source of inspi-

ration. The study was supported by a grant from the Gustav and Louise Pfeiffer Research Foundation of New York City and the China Medical Board.

Materials and methods

General Description. — A detailed description of *Bufo melanostictus* SCHNEIDER from the Indo-Australian Archipelago is given by VAN KAMPEN (1923) and need not be repeated here. Perhaps because of its distinctive black crests, *melanostictus* is easily identified and rarely if ever confused with any other species. As is usually the case in *Bufo*, the parotoid glands are large and in this species, more-or-less oblate; the back and sides are conspicuously covered with black warts. Occasionally an animal is found with white or yellowish crests and warts, but this has been ascertained to be a transient condition that occurs when the skin is shed. A persistent characteristic in the species on Java and Bali, but which seems to have been overlooked in the descriptions given by BOULENGER (1912) and VAN KAMPEN (1923) is the presence of two large black warts located close together on the dorsal median line just above the juncture of the cranium with the first vertebra. This landmark is extremely useful in pithing animals as it indicates the point at which the needle should be inserted.

The parotoid glands and skin exude a sticky mucus-like substance which, after handling many animals, covers one's hands and is difficult to remove. In spite of the fact that there is a wide-spread belief throughout Java and Bali that toads will produce various harmful effects if touched, nothing untoward was noted during this study which might have been attributed to the effects of this secretion. NOBLE (1931) reports that the secretions of *Bufo* cause serious inflammation of the eyes of higher vertebrates, but on two occasions the parotoid secretions of *melanostictus* were inadvertently introduced into the eyes of the author without ill effect. Nevertheless, these glands no doubt provide some protection, perhaps by producing an unpleasant taste, because dogs and cats and other domesticated animals found around human habitations leave them alone. Snakes, however, are known to devour *melanostictus* without being harmed.

The color of *melanostictus* on Java and Bali is extremely variable, ranging from yellow through red to black. At night they tend to be lighter in color or yellow with black warts, frequently speckled with gray or mottled with red. The same animals in the daytime, however, sometimes become darker and frequently appear to be gray or black. Males in am-

exes have often been noted to be lighter in color than usual. Secondary x characteristics are marked in males and they can always be easily identified by the redness of the skin under the jaw, and by the black nuptial pads on the first two fingers (CHURCH & THE, 1958). The third finger on occasional specimens exhibits faint pigmentation, but it is not the general rule as in China (LIU, 1936). The redness found under the jaw is gradually lost when the animals are preserved in formalin. Data for all are incomplete, but on Java the males are known to display secondary x characteristics at all times of the year.

Ecological Notes. — With the exception of four localities, the animals were all collected at sea level. Bandung, however, is eight hundred meters above sea level; Sukabumi is six hundred; Malang is four hundred; and Bogor is three hundred. There are undoubtedly variations in the daily temperature ranges between these four cities and those at sea level, and the growth of the animals in these four regions may have been affected by the lower temperatures. However, no data at present available would show this to be so. Although Bogor receives more rainfall than other parts of Java, the conditions of humidity and precipitation in the sixteen localities are comparable. Presumably there is a constant food supply available. Stomachs of animals examined from the Djakarta area showed a continuous supply of food throughout the year, the contents consisting mainly of insects and the grubs of beetles (CHURCH, 1958).

The local habitats of this species in each town where they were collected were similar, if not identical. Open places, especially without grass such as school yards in which night lights were burning, open areas around among houses, along the streets and under the street lights, open playing fields, under trees, amongst trash and decaying vegetation, in gardens, and in some cases, even in houses, offered the most likely places to find them. They were rarely found in flooded rice paddies, on swampy ground or along the edges of rivers or streams, nor did they appear to inhabit extensively fields with tall grass. They were frequently found in drainage ditches in standing or slow-moving water, and in Solo a large number of males were collected around the pond in the zoological garden. They obviously prefer areas of human habitation.

The preference of *melanostictus* for a particular habitat was most clearly demonstrated in the lowlands just north of Djakarta between Keajoran and Tandjung Priok. The area is given over entirely to rice swamps except for native kampongs set in it like islands. These oases of human habitation are about one kilometer apart and are connected by

paths through the flooded sawahs. When collecting in this area, few if any *melanostictus* were encountered in the sawahs although there were many *Rana*, but they occurred in great abundance in the open, grassless spaces around the houses of the kampongs where they enjoyed a free range unhindered by any other species.

A similar tendency on the part of *melanostictus* to occupy a particular habitat was also found to be the case in Jogjakarta and especially in Surabaya where these toads compete with *B. biporcatus* for *Lebensraum* (CHURCH, 1959). Unlike West Java, where *biporcatus* is rarely found, (only ten or so were found in collection of over two thousand animals taken around Djakarta), the two species in Jogjakarta and Surabaya exist in almost equal numbers. But though they were found in closely neighboring areas, *biporcatus* preferring — perhaps not by choice but by necessity — the more moist sawahs and river banks and *melanostictus* the open areas around buildings, they were not often found together. Evidence from East Bali, however, where *melanostictus* apparently does not occur, the area thereby being given over to *biporcatus*, demonstrates that *biporcatus* will also invade the habitat preferred by *melanostictus* if *melanostictus* is not present. In Ubud and Den Pasar, East Bali, for example, *biporcatus* was observed in great abundance not only in their usual moist habitats but also in gardens and around houses, the typical places to expect to find *melanostictus*. The same was also true at Karang Asem, a town on the easternmost tip of Bali. On the other hand, in Negara, West Bali, where *melanostictus* has presumably recently been introduced (CHURCH, 1959), *melanostictus* has become the dominant species of the two and has pushed *biporcatus* aside, reducing its numbers as compared with East Bali, and creating the same conditions of separated local habitats for the two species as exist in Jogjakarta and Surabaya.

Collecting Notes. — A total of 3,908 *melanostictus* were used in this study. The majority of the specimens were collected by the author and his assistants. Those from the two Djakarta areas, Menteng and Kemajoran, which represent the two largest series, were collected over a period of a year, from September, 1957, to September, 1958. Those from Kampong Kamang, a rural area about ten miles south of Djakarta, were collected over a period of three months, from December, 1957 to February, 1958. All of these animals were utilized in other experimental studies which led to considerable mutilation, so they were not preserved.

The specimens from Serang, Pelabuhan Ratu, Solo, Surabaya, Malang, Madjalengka, Pamekasan (Madura), Banjuwangi and Negara (Bali) were

collected in one evening in each town. Those from Jogjakarta were collected on three successive evenings, and those from Bogor were collected over a period of weeks. The animals from Sukabumi and Bandung were collected on different occasions two or more months apart. Except for the animals from Bandung and Madjalengka which were kept alive for forty-eight hours after capture, all of the animals were killed within twenty-four hours after being collected, and the snout-vent measurements made with vernier calipers to the nearest millimeter before being preserved in formalin.

The stage of the moon affected the collections somewhat. During the time of the full moon, more females than males were found. And when there was no moon, the males were more abundant. In Solo, in one evening when there was no moon, 107 males and 23 females were collected. Collecting notes from Ubud, Bali, during a two-week period when the moon was waxing, revealed a gradual increase in the number of *B. biporcatus* females caught every evening, until finally the ratio of males to females was about 50-50. Something similar can be said of certain species of *Rana*. These findings, which are in keeping with the rhythmic ovulation cycle described for *melanostictus* by CHURCH (1958), would also indicate that the lunar cycle may exert some influence on all amphibians in this area.

Results

The statistical data pertaining to the snout-vent measurements made on the sixteen series of *B. melanostictus* are summarized in Table I. A recapitulation of the entire collection is also given.

TABLE I. VARIATION IN SNOUT-VENT MEASUREMENTS OF SIXTEEN POPULATIONS OF *Bufo melanostictus* FROM JAVA AND BALI. THE FINAL COLUMN REPRESENTS WHETHER OR NOT THE DIFFERENCE BETWEEN THE MEAN OF A POPULATION AND THAT OF THE STANDARD IS SIGNIFICANT

Place	Sex	Number of Animals	Range	Mean \pm SE	+ = significant dif. — = not significant
Serang	M	52	38 — 83 mm.	62 \pm 1.01	
	F	68	44 — 89 mm.	64 \pm 1.14	
	M+F	120	38 — 89 mm.	63 \pm .77	+
Menteng (Djakarta)	M	276	50 — 86	68 \pm .41	
	F	808	50 — 95	71 \pm .28	
	M+F	1,084	50 — 95	70 \pm .24	—
Kemajoran (Djakarta)	M	148	59 — 95	73 \pm .50	
	F	612	50 — 98	76 \pm .40	
	M+F	760	50 — 98	76 \pm .34	+

Place	Sex	Number of Animals	Range	Mean \pm SE	+ = significant dif. - = not significant
Kampong Kamang	M	115	65 — 101	79 \pm .58	
	F	194	59 — 104	89 \pm .54	
	M + F	309	59 — 104	85 \pm .49	+
Bogor	M	89	35 — 95	72 \pm 1.39	
	F	85	38 — 107	77 \pm 1.94	
	M + F	174	35 — 107	75 \pm 1.18	+
Pelabuhan Ratu	M	9	56 — 80	70 \pm 2.70	
	F	19	44 — 98	75 \pm 2.93	
	M + F	28	44 — 98	73 \pm 2.20	—
Sukabumi	M	90	38 — 83	64 \pm 1.10	
	F	125	35 — 89	69 \pm 1.00	
	M + F	215	35 — 89	67 \pm .76	+
Bandung	M	78	53 — 83	69 \pm .81	
	F	101	47 — 92	70 \pm 1.03	
	M + F	179	47 — 92	70 \pm .75	—
Madjelengka	M	77	50 — 86	69 \pm .81	
	F	20	62 — 98	74 \pm 2.06	
	M + F	97	50 — 98	70 \pm .79	—
Jogjakarta	M	72	56 — 92	74 \pm .92	
	F	64	47 — 110	77 \pm 2.00	
	M + F	136	47 — 110	76 \pm 1.06	+
Solo	M	110	56 — 83	70 \pm .53	
	F	32	44 — 89	64 \pm 1.03	
	M + F	142	44 — 89	69 \pm .64	+
Surabaya	M	36	59 — 83	72 \pm .91	
	F	62	47 — 92	71 \pm 1.40	
	M + F	98	47 — 92	72 \pm .95	—
Malang	M	27	53 — 86	66 \pm 1.63	
	F	63	44 — 89	62 \pm 1.43	
	M + F	90	44 — 89	63 \pm 1.20	+
Madura	M	46	47 — 74	63 \pm .82	
	F	111	38 — 86	60 \pm 1.10	
	M + F	157	38 — 86	61 \pm .82	+
Banjuwangi	M	59	44 — 77	61 \pm .81	
	F	109	41 — 86	59 \pm 1.03	
	M + F	168	41 — 86	60 \pm .75	+
Negara, Bali	M	71	56 — 80	66 \pm .55	
	F	80	59 — 92	76 \pm .77	
	M + F	151	56 — 92	71 \pm .62	—
* * * *					
Entire Collection	M	1,355	35 — 101	69 \pm .24	
	F	2,553	35 — 110	72 \pm .24	
	M + F	3,908	35 — 110	71 \pm .18	

As can be seen from Table I, the range in size of females generally exceeds that of males. Nevertheless, the average sizes for males and females are almost the same in all but three populations. Only the series from Kampong Kamang, Solo and Negara show a statistically significant difference between the means of the two sexes. However, considering the small number of females in the Solo series, the difference between the means of the males and females is probably not accurate, and measurements on a larger number of females would no doubt show them to be on the average about the same size as the males.

When the means for the entire collection are considered, the similarity in average size between the two sexes becomes even more marked. The average snout-vent size for all of the males and all of the females together is 71 mm; that for all of the males is 69 mm; and for all of the females, 72 mm. Even though females are found which are larger than the largest males, the findings demonstrate that there is no marked sexual dimorphism in this species as far as snout-vent size is concerned on Java, as there is on Bali, and as LIU (1936) reports to be the case in China. It is interesting to note, however, that the author has never found a male in amplexus with a smaller female. In mating pairs the female is invariably the larger, and were one to compare the sexes by this criterion alone, one would be led to assume that a marked sexual dimorphism did indeed exist in size.

The largest male measured 101 mm; the largest female measured 110 mm. One enormous female which measured 120 mm was collected in Jogjakarta, but it was not included because its extreme size put it in a class by itself.

Both the smallest male and the smallest female measured 35 mm. Whether animals of this size are capable of mating is unknown. Viable sperm have been found in the testes of males of this order (CHURCH & THE, 1958), but the smallest female found with mature eggs in the ovaries measured 40 mm (CHURCH, 1958).

The means of the snout-vent lengths of the sixteen populations varied from 60 mm in Banjuwangi to 85 mm at Kampong Kamang (Djakarta). Taking the average snout-vent measurement of 71 mm for the entire collection as a standard, 10 populations showed differences between the means of more than three times the sum of the standard errors which would indicate different populations. A comparison of the means of the different series with the mean of the entire collection is shown graphically in fig. 1.

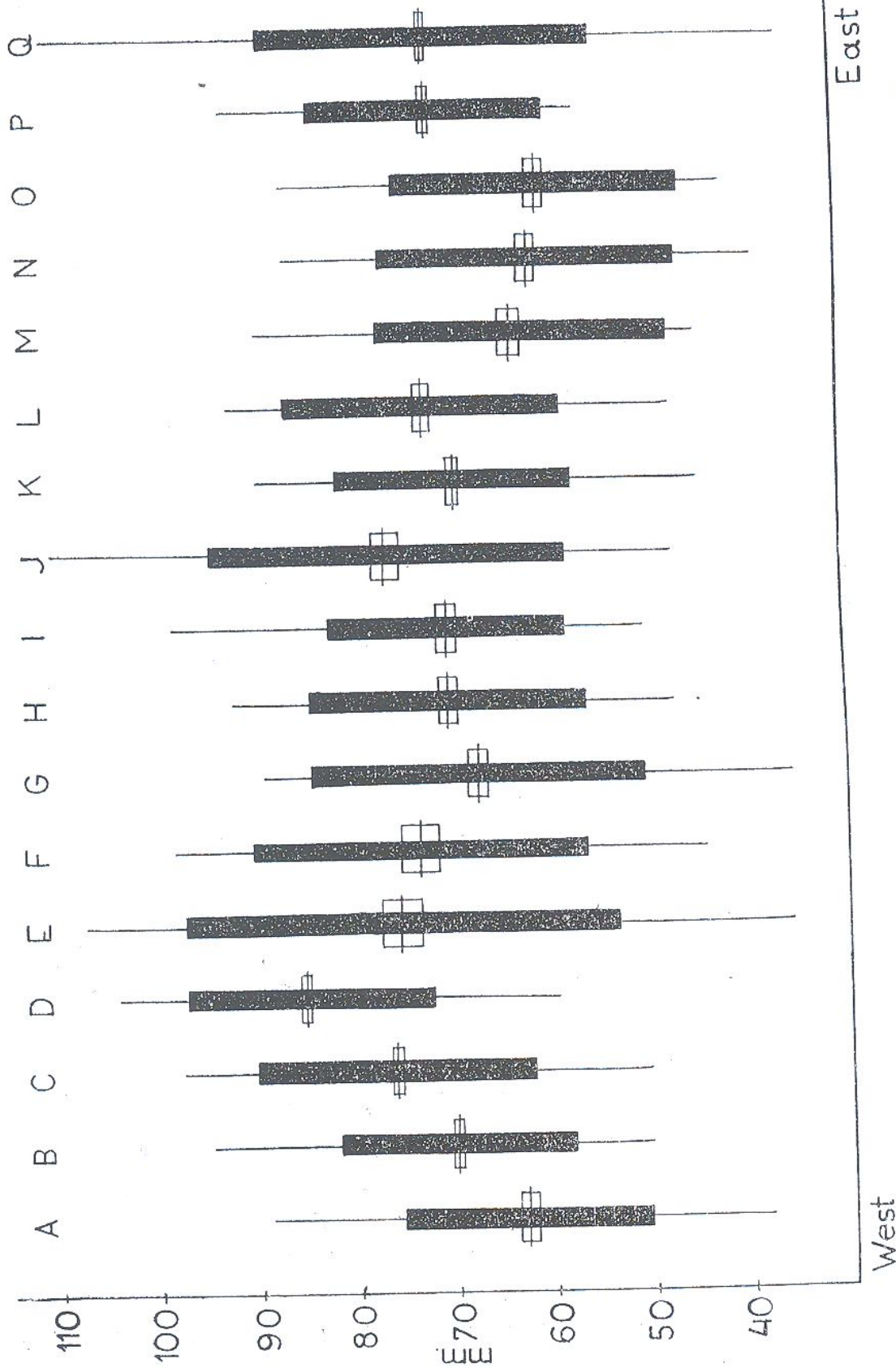


FIG. 1. Variation in snout-vent length in sixteen populations of *Bufo melanostictus* on the islands of Java and Bali. The vertical lines represent the size ranges; the vertical bars, three standard deviations; the horizontal lines, the means; and the horizontal bars, two standard errors. A, Serang; B, Menteng (Djakarta); C, Kemajoran (Djakarta); D, Kampong Kamang; E, Bogor; F, Pelabuhan Ratu; G, Sukabumi; H, Bandung; I, Madjalengka; J, Jogjakarta; K, Solo; L, Surabaja; M, Malang; N, Madura (Pamekasan); O, Banjuwangi; P, Negara, Bali; Q, Entire collection.

Animals from populations with means which were significantly larger than the overall average size of 71 mm were found to occur in smaller numbers and they were on the whole more active than their smaller counterparts from other areas. Conversely, in areas where the mean of the populations was found to be smaller than the overall average size of 71 mm, the toads were present in great abundance and were very easy to collect. In Jogjakarta, for example, where the average size for the species was found to be 76 mm, it took about six hours on two successive evenings to collect 50 animals. On the first evening it was dry, and in the afternoon of the second day there was a heavy rain. However, animals were generally difficult to find on both evenings which may have been due in part to the increased activity of that particular population.

On the other hand, in Banjuwangi where the average size turned out to be 60 mm, 168 animals were collected in about an hour and a half in one evening. The habitats, temperature and weather conditions of both places were comparable, but the smaller toads of Banjuwangi were not so active and were much easier to catch than those from Jogjakarta. Interestingly enough, blood studies being carried out on this species indicate that the hemoglobin concentration is higher in animals from populations in which the size is generally larger; this finding might in part account for the increased activity noted in the larger-sized animals.

Data on the occurrence of vocal slits in the males were collected on thirteen populations. They are listed in Table II. Usually there was only one opening into the internal median vocal sac, either to the right or to the left of the median line in the floor of the mouth. Taking the collection as a whole, 38% of a total of 886 males had the opening on the right; 46% had the opening on the left; 11% had both right and left openings; and 5% had none. A comparison with position of vocal slits in males from China is also given in Table II.

TABLE II. POSITION AND PERCENTAGE OF VOCAL SLITS IN MALE *Bufo melanostictus* FROM DIFFERENT POPULATIONS

	Right	Left	Both	None	Total number
Serang	37%	44%	13%	6%	52
Menteng	40	51	5	4	163
Kemajoran	41	44	5	10	78
Bogor	33	44	6	17	84
Sukabumi	35	44	10	11	63
Bandung	40	49	10	1	43

TABLE II (continued)

	Right	Left	Both	None	Total number
Jogjakarta	27	55	16	2	74
Surabaja	44	33	20	3	36
Solo	44	44	12	0	107
Pamekasan	43	39	18	0	46
Malang	33	33	27	6	26
Banjuwangi	34	42	20	4	59
Negara (Bali)	38	48	5	9	56
China (Liu, 1936)	29	36	27	8	55

Discussion

The results of this study indicate that considerable variation exists in snout-vent size between local populations of *B. melanostictus* on Java and Bali. Probably further variations would be found if comparisons could be made with populations on other islands of the Archipelago.

If the mean snout-vent length of 17 mm for the entire collection is taken as a standard, significant differences can be shown to exist between this standard and ten of the sixteen series (Table I). However no population difference is of an order to suggest subspecific distinctness. The population sample from Kampong Kamang, for example, whose mean differs most widely of all from the standard of 71 mm, shows an overlap of about 20% with the standard population. This difference might be taken to indicate separate populations, but it is not great enough to warrant consideration of the Kampong Kamang animals as a subspecies.

It is doubtful whether these variations can be attributed to differences in temperature and humidity at the various localities, especially not for those populations at sea level. There are known variations in the richness of the soil throughout Java which might produce variations in nourishment, but the available soil research information cannot be correlated with the size differences of *melanostictus*. Jogjakarta represents an area with good soil, as does Banjuwangi; the temperature and humidity of both places are comparable. If the soil were to affect the quality of the nutrient intake to such an extent as to alter the size of animal populations, its effects should be the same on all populations living in similar areas. Yet in Jogjakarta the animals were found to be significantly larger than the average standard, and in Banjuwangi they were significantly smaller.

Nor can competition with another species for food and living space explain the size differences in different populations. One would expect that such competition, by reducing the food supply, would reduce the growth. However, the only other species found in large numbers in allopatric contact with *melanostictus* is *B. biporcatus*. In Jogjakarta, Surabaya and Negara where the two species both were found in more-or-less equal numbers, *melanostictus* was average or larger in size; whereas in Banjuwangi, Madura and Serang — places where no *biporcatus* were found — *melanostictus* was smaller than average.

No dispersal pattern of the species throughout Java is made readily apparent by the findings. Hypothetically, the species arrived in the Archipelago from Malaya; and in Java, from Sumatra. If this is so, it could most easily have migrated eastward in Java along the northern coastal plain. Further dispersal southward could then have been effected via the low-lying regions between the mountains of the central range. Considering the preference of this species for areas around human habitations, its dispersal may have been further influenced by this factor.

There is some evidence, though inconclusive, that migration along the well-populated northern coastal plain may have occurred. The animals collected at Menteng (Djakarta), Madjalengka and Surabaya all have an average size of about 71 mm. The series collected at Negara, Bali, might also belong to this group. Applying the test for population variation which shows two or more sample populations to be different if the difference between the means of the samples is more than three times the standard errors of the difference, the above four series can be shown to be comparable in size and perhaps of the same continuous population. The twelve populations to the north and to the south of the coastal plain might be considered as off-shoots from this main line of migration (fig. 2).

It is known that *melanostictus* does not occur at Ujung Kulon, the game preserve at the southwestern tip of Java (WEGNER, 1958), and that again would indicate that the species migrated along the northern rather than the southern coast. But further collections along the at-present inaccessible and less-densely populated southern coast of Java are needed to help complete this picture. Information of the snout-vent size of *melanostictus* on the southernmost tip of Sumatra would also be important. What few measurements of the species there are available from Sumatra show no appreciable difference when compared to the overall average of the animals on Java, but it would be interesting to ascertain specifically if the small size recorded at Serang on the western tip of Java is comparable to that of the toads in southern Sumatra. At the other end

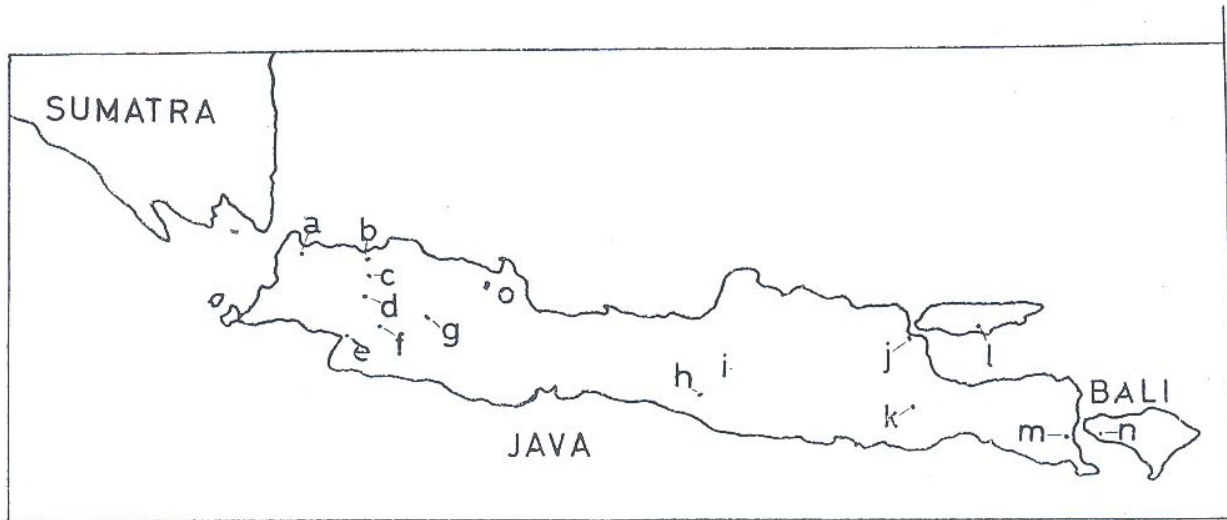


Fig. 2. Map of Java and Bali showing where *Bufo melanostictus* were collected. a, Serang; b, Djakarta (Menteng, Kemajoran); c, Kampong Kamang; d, Bogor; f, Sukabumi; e, Pelabuhan Ratu; g, Bandung; o, Madjalengka; h, Jogjakarta; i, Solo; j, Surabaya; k, Malang; l, Pamekasan; m, Banjuwangi; n, Negara.

of Java, between Banjuwangi and Bali, such is not the case, the *melanostictus* of Bali being larger than the animals found at the westernmost extremity of Java in Banjuwangi. Surabaya is the nearest known point in Java with a population comparable in snout-vent size to that of Negara, and it is of course possible that they could have arrived in Bali from there.

It has been suggested by CHURCH (1959) that *melanostictus* is a relative new-comer to the Sunda Islands and that it is gradually displacing *B. biporcatus* as it moves eastward into the Archipelago. Evidence supporting this suggestion can be found in the limited range of *melanostictus* which reaches only the westernmost tip of Bali as compared to *biporcatus* which is found as far as Lombok, the presence of *melanostictus* and the absence of *biporcatus* in Malaya, and the gradually increasing density of the *biporcatus* population from west to east in Java suggesting displacement as *melanostictus* migrates eastward. If *melanostictus* is indeed invading a biotope already occupied by *biporcatus*, this factor, too, could hinder and influence its dispersal.

It has often been pointed out that the absolute size of poikilothermous animals which continue to grow throughout their lives is extremely variable and this fact has been borne out by this study. The range in snout-vent length presented here is from 35 to 110 mm. When the wide range in size is considered, the accuracy of analyses of small samples can be opened to question. The sample of the Solo population most clearly demonstrates this point. Here a larger number of males than females was collected. The size ranges of the two sexes turned out to be comparable

with those of other areas, but because of the smaller number of female specimens available, the mean snout-vent size of the female was significantly smaller than that of the males. Obviously, further collections in the Solo area would demonstrate the size of the females there to be comparable to, or larger than, that of the males. When even smaller numbers of specimens are analysed, in spite of the application of statistical methods of correction, it is the opinion of the writer that the results can be only misleading.

The problem of species size is also made difficult by population studies of continuously growing animals. Both BOULENGER (1912) and VAN KAMPEN (1923) have reported the maximum size of *melanostictus* in Malaya and the Archipelago to be 115 mm. But when one considers that in this study only one animal out of 3,909 animals collected over an area of 100,000 square kilometers exceeded 110 mm in snout-vent length, the practice generally in use of reporting only the size of the largest specimen of a species collected turns out to be highly inaccurate and misleading as far as practical purposes are concerned.

On the other hand, upholding the greater accuracy of larger population samples, it is interesting and somewhat curious to note that determinations of erythrocyte numbers and hemoglobin concentrations made on the Djakarta populations of *melanostictus* (CHURCH, TIO & KUSIN, 1958) when correlated with snout-vent length, indicate the optimum erythrocyte number and hemoglobin concentration to fall at the mean of the population sample. Since different animals were used for the snout-vent study and for the blood studies, the blood determinations would offer confirmation of the accuracy of the average sizes obtained in these population samples.

Summary

1. The results of analyses of snout-vent measurements on 3,908 *Bufo melanostictus* SCHNEIDER collected in 16 localities on Java and Bali show that considerable variation exists in size between local populations of this species in these areas.

2. The range in size of females exceeds that of males, but the average sizes for males and females were found to be almost the same in all but three of the sixteen populations.

3. The average size for the whole collection was 71 mm. The largest male measured 101 mm, the largest female measured 110 mm. The smallest male and the smallest female measured 35 mm.

4. Using the average snout-vent measurement of 71 mm as a standard, 10 populations showed statistically significant differences between the means.

5. Animals from populations which exceeded the standard of 71 mm were found to be more active than those from populations which were smaller than the standard.

6. Data on the occurrence of vocal slits in 886 males were collected from 13 populations. 38% were found to have one vocal slit located on the right side; 46% had one vocal slit on the left; 11% had both right and left vocal slits; and 5% had none.

7. A discussion of the possible factors causing size variation, and the possible route of dispersal of this species in Java and Bali is given.

REFERENCES

- BOULENGER, G. A., A Vertebrate Fauna of the Malaya Peninsula, Taylor and Francis, London, 1912.
- CHURCH, G., 1958. The reproductive cycle of the Javanese toad, *Bufo melanostictus*. Part I. Ovulation in *Bufo melanostictus*. Indonesian Council for Sciences.
- , 1959. The invasion of Bali by *Bufo melanostictus*. In preparation.
- and THE DJIN TEK, 1958. The reproductive cycle of the Javanese toad, *Bufo melanostictus*. Part II. Cyclic changes in the testes of *Bufo melanostictus*. Indonesian Council for Sciences.
- and JANE A. KUSIN and TIO KIAN LING, 1958. The reproductive cycle of the Javanese toad, *Bufo melanostictus*. Part IV. Cyclic changes in the hemoglobin concentration and the number of erythrocytes in *Bufo melanostictus*. Indonesian Council for Sciences.
- KAMPEN, P. N. van, The Amphibia of the Indo-Australian Archipelago. Leiden, 1923.
- LIU, C. C., 1936. Secondary sex characters in Chinese frogs and toads. Field Mus. Nat. Hist., Zool. Ser., 22: 115.
- , 1950. Amphibians of Western China. Fieldiana: Zool. Mem., vol. 2.
- NOBLE, G. K., The Biology of the Amphibia. McGraw-Hill, New York, 1931.
- SMITH, M. A., 1930. The Reptilia and Amphibia of the Malay Peninsula. Bull. Raffles Mus., No. 3.
- WEGNER, A. M. R., 1958. Personal communication.