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Michael J. D. White (Photograph by David Lamb-Jenkins)
Toward a genodynamics of hybrid zones: studies of Australian frogs and West Indian land snails

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The biological species concept is a central tenet of evolutionary biology. Hybrid zones of various kinds threaten this concept by revealing that species may lack some of the properties (integrity, cohesion, and reproductive isolation) that we have ascribed to them. Whereas hybrid zones have traditionally been treated as taxonomically bothersome (but not unexpected) phenomena involving borderline cases, they are increasingly being regarded as evolutionists' delights that allow us to monitor the processes of geographic differentiation and ultimately speciation. In my view the elucidation of the roles of gene flow and natural selection in a natural hybrid zone is a prerequisite to the solution of the problems presented by that zone. On a more general level, the need to develop empirical techniques to measure the genodynamics of hybrid zones is critical if we are to understand the processes by which speciation occurs and the entity of the species itself. Until we learn more about what is going on in nature, it strikes me that assertions that the species is not the keystone of evolution are simply premature. Whether species are "mental abstractions which order clusters of diversity in multidimensional character space" (Levin, 1979:381), or whether they have reality, "a reproductive community, a gene pool, and a genetic system" (White, 1978), remains to be seen. Obtaining the answers will certainly require a more rigorous application of the techniques of population biology and genetics; we can no longer entrust these important phenomena exclusively to Darwin's (1859) "naturalists having sound judgement and wide experience."

In this chapter, I will illustrate the extent to which our present theory of hybrid zones is inadequate by describing two groups of animals in which hybridization is a prominent feature. I will point out the nature of the problems encountered in each group and note the extent to which these problems might be alleviated if we understood the genodynamics of the
interactions. I will suggest ways in which existing techniques might be applied to these problems. Although no single hybrid zone seems amenable to the full range of investigations that should be conducted, it seems likely that we can make substantial progress by conducting more detailed analyses of a few selected cases.

At the outset it should be noted that much of the difficulty surrounding the interpretation of natural hybrid zones stems from the fact that the “solution” of the problem of each particular interaction really involves the solution of four separate questions:

1. What is the geographical relationship of the ranges of the interacting populations?
2. What is the history of the interaction?
3. What will be the probable outcome of the interaction?
4. What is the taxonomic status of the interacting population?

Usually only the first of these questions can be answered objectively. To answer the second question one must be able to distinguish between cases of primary and secondary intergradation, a distinction that is usually impossible to make. The answer to the third question stems from detailed studies of the interacting taxa in nature and the laboratory. Our predictive theory is still so inadequate, however, that even when such information is available, it is only rarely that the third question can be answered. Logically, the answer to the fourth question depends on the answers to the previous ones. Yet, in practice, it has often been the first to be “solved.” Many of the older controversies involving particular cases of hybridization arose because undue emphasis was placed on the premature solution of the final question.

A second source of confusion must also be noted; several of the words used to describe hybrid phenomena are actually nonoperational because they are defined in terms of “answers” to the second, third, or fourth questions. I have proposed new criteria for describing cases of natural hybridization that overcome the difficulties inherent in schemes based on the history of the interaction (primary or secondary contact) or the taxonomic status of the interacting forms (Woodruff, 1973a). In this paper I will employ the terms allopatric and parapatric hybridization to imply geographical information only.

**Pseudophryne hybrid zones in southeast Australia**

*Pseudophryne semimarmorata, P. bibroni, and P. dendyi* are small leptodactylid frogs with generally contiguous distributions in southeast Australia (Figure 9.1). Narrow zones of parapatric hybridization have been found wherever the northern border of *P. semimarmorata*
comes into contact with the southern borders of P. bibroni and P. dendyi. These hybrid zones are particularly interesting because they provided direct evidence for the operation of postmating isolating mechanisms in animals in a natural situation (Woodruff, 1972b). They are also of special interest in that they involve anurans whose breeding calls are indistinguishable and therefore may not serve as premating isolating mechanisms.

A highly abridged summary of the following account has appeared elsewhere (Woodruff, 1979).

The *Pseudophryne*, commonly called toadlets, differ from typical anurans in a number of ways. They are terrestrial and cryptozooic as adults. They lack a tympanum, middle ear, and expansive vocal sac, and the breeding call is practically identical in all members of the genus. In the three species under consideration breeding is restricted to the autumn months. Several weeks before mating, males migrate to the breeding area and commence vocalization. In southern Victoria these breeding choruses of 20–50 males form in the last third of March in most years. Males maintain territories and prepare nest sites among the grass roots and leaf litter in situations that will be saturated with water during the winter. Females enter the area and begin mating in the last third of April; they lay 70–90 large eggs in discrete batches and may mate on several occasions over a 4–7-week period. After mating a male usually remains with the eggs and resumes calling, mating again if the opportunity occurs. The embryonic phase of development is protracted, and although hatching

Figure 9.1. Map of southeast Australia and Tasmania showing the general ranges of three species of *Pseudophryne.*
normally occurs after 4–6 weeks, it may be delayed until conditions are favorable for the aquatic larval stage. Metamorphosis occurs in September and October, and toadlets are thought to reach maturity in 2 or 3 years. Adults of these three species have consistently different patterns of coloration. They are, in contrast, very similar in anatomy, karyotype, size, breeding season and site, common male vocalization, pre- and postmating behavior, reproductive rates, mating system, and the pattern and rates of embryonic and larval development. I have supported the above generalizations elsewhere (Woodruff, 1972a, b, 1975c, 1976a, b, 1977, 1978a).

A zone of parapatric hybridization stretches discontinuously over 750 km along the northern border of P. semimarmorata (Littlejohn, 1967; Woodruff and Tyler, 1968; Woodruff, 1979). Five diagnostic adult coloration characteristics were used to distinguish the taxa, estimate the hybridity of toadlets of intermediate coloration, and map the hybrid zones.

Figure 9.2. Morphological variation of *Pseudophryne* near Wallan: Hybrid index diagrams illustrate the change from *P. bibroni* (locality A and X) to *P. semimarmorata* (locality G and Z). The hybrid indexes range between 0 (typical *P. semimarmorata*) and 10 (typical *P. bibroni*). The diagrams show all specimens collected at selected localities between 1960 and 1969. The 306-meter and 350-meter (shaded) contour lines are indicated; www is the Wandong-Whittlesea-Woodstock road junction, 3 km east of Wallan.
Detailed sampling in seven areas revealed that the interactions between *P. semimarmorata* and *P. dendyi* and between *P. semimarmorata* and *P. bibroni* were very similar in many respects. Morphological variation in two of these areas is summarized in Figures 9.2 and 9.3. The hybrid zone is generally less than 9 km wide, and over 80% of the morphological change occurs in the central 3 km. The diversity of hybrid phenotypes suggests that mating probably occurs at random among toadlets at each breeding site. No evidence for assortative mating based on ecological, ethological, or morphological differences was detected. There are slight asynchronies in the breeding seasons of the interacting taxa, but they do not appear to limit hybridization during the middle 80% of the season. Postmating isolating mechanisms are not sufficient to prevent the occurrence of apparently healthy *F₁* and backcross hybrids. In vitro hybridization experiments and studies of field-collected eggs confirmed that whereas hybrids are both viable and fertile, hybrid embryos are less viable than homospecific ones.

Figure 9.3. Distribution and morphological variation of *Pseudophryne* from the hybrid zone between *P. semimarmorata* and *P. dendyi* near Tyers. The map shows the position of collecting sites in relation to roads (heavy lines) and villages. The hybrid index diagrams describe variation at 12 selected localities (A–L): Hybrid indexes range between 0 (typical *P. semimarmorata*) and 10 (typical *P. dendyi*). The fine contour lines are isophenes connecting areas of similar mean hybridity.
Although natural embryonic mortality rates away from the hybrid zones are typically less than 5% (range 0–10%), many batches of eggs collected from within the hybrid zone suffered levels of mortality two to three times as high (Woodruff, 1972b, 1979). The range of embryonic mortality values seen in field-collected eggs within the hybrid zones indicates that some recombinations may be less viable than others (Figure 9.4).

The histories of these interactions are conjectural. Littlejohn (1967) and Littlejohn and Martin (1974) interpreted them as the outcome of secondary contacts between taxa that diverged from one another in allopatry. Littlejohn suggested that P. dendyi was derived from a P. bibroni-like ancestor during the last glacial period when it became isolated to the south of the Eastern Highlands. Littlejohn and Martin suggested that P. semimarmorata was also derived from the same ancestral stock, but that its differentiation occurred when it became isolated on the continental island of Tasmania during interglacial times. According to these workers, Pseudophryne reached Tasmania by way of the land bridge that emerged during the penultimate glaciation and returned to the mainland as P. bibroni.

Figure 9.4. Elevated levels of embryonic mortality were associated with batches of eggs collected in Pseudophryne hybrid zones. The P. semimarmorata–P. bibroni hybrid zone near Wallan is shown on the left, the P. semimarmorata–P. dendyi hybrid zone near Tyers on the right. The upper diagrams show the mean and range of hybrid index scores of samples from localities described in histograms in Figures 9.2 and 9.3. The lower diagrams show embryonic mortality (mean values connected for the Wallan case) in batches of eggs collected at these localities. (After Woodruff, 1979)
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*semimarmorata* on a second land bridge that emerged during the last glacial phase. According to these scenarios the present hybrid zones could be 10,000–20,000 years old. An alternative scenario involves the hills of the Eastern Highlands as the barrier that facilitated the differentiation of both the southern toadlets. I have argued (Woodruff, 1972b) that present patterns of distribution and morphological variation in *P. semimarmorata* and *P. dendyi* suggest that neither *P. dendyi* nor *P. bibroni* was present in southern Victoria at the various times Tasmania was connected to the mainland. Although the details of the evidence supporting this position no longer seem important, it should be noted that if the highlands rather than Bass Strait served as the barrier facilitating allopatric differentiation of *P. semimarmorata*, then again the present hybrid zone may be at least 10,000 years old.

Two other hypotheses must also be considered. McDonnell et al. (1978) have proposed that the zones (specifically the Wallan contact) may be of more recent origin. Arguing that agricultural activities have increased the area of suitable habitat for *Pseudophryne*, they concluded that such changes would have hastened contact or intensified any existing interactions. It is my subjective impression (based on over 70 field trips) that breeding choruses at undisturbed sites may, in fact, be smaller than those associated with roadside ditches. It seems quite probable that human activities during the last 100 years have had a considerable effect on the local abundance of these animals. That such activities had a marked effect on their overall distribution seems less likely though, for man has tended to modify the habitats originally favored by *Pseudophryne* rather than those where it is not found today even in undisturbed situations. I suspect the hybrid zones may antecede the arrival of Europeans in the region.

A radically different interpretation of these interactions has been offered by Endler (1977), and I have discussed it elsewhere (Woodruff, 1978b, 1979). Although not refuting the allopatric models, Endler has shown how the zones could equally well have evolved by parapatric differentiation. According to this view, they represent complex step clines separating areas characterized by different coadapted modifiers that interact so as to reduce the fitness of the hybrids. The hybrid zone effect is therefore the result of clinal selection patterns between the slightly different habitats of the interacting taxa. The taxa themselves probably differ in relatively few major genes (some of which control coloration); it is the milieu of these genes that is important. In this scenario the present position of the zones may be related to present environmental gradients and partial barriers—which affect population density, to the slopes of the
selection gradients, and to the history of these variables. Endler's contribution is most important because he has shown how available techniques do not allow us to distinguish between the results of allopatric and parapatric differentiation. Although the various historical hypotheses are all plausible, they are untestable at present and therefore outside of science; the origin of the zones cannot be established yet.

Perhaps we can make some progress by exploring the dynamics of the zones today. If an adequate model can be developed for the existing phenomena, then we may gain new insights into the past and possibly even the future. Three major features of these hybrid zones must be accounted for:

1. There is extensive hybridization between individuals of dissimilar allopatric populations in a restricted narrow zone whose present position is not clearly related to any single environmental factor.
2. Despite the extensive interbreeding and backcrossing, introgression into either homospecific population is apparently very limited.
3. The zone is relatively stable, there being no evidence that fusion is occurring or that premating isolating mechanisms are being reinforced.

One hypothesis to account for these features is suggested by a closer consideration of the intrinsic isolating mechanisms that may operate between these interacting taxa. Let us first review the potential premating isolating mechanisms. No ecological differences were detected that might serve as isolating mechanisms; the interacting taxa appear almost identical in their requirements in the areas where their ranges meet. No morphological differences that might constitute isolating mechanisms were noted; these taxa are very similar in size and shape, and the differences in adult coloration are not visible at night when the toadlets typically mate (Woodruff, 1972b). Behavioral differences were likewise judged to be insignificant. Acoustic signals (key isolating mechanisms in many anurans) are not differentiated. McDonnell et al. (1978) studied breeding calls near the \textit{P. semimarmorata-}\textit{P. bibroni} contact at Wallan and found overlap in all seven call characters investigated. Pengilley (1971) found the breeding calls of \textit{P. bibroni} and \textit{P. dendyi} to be so similar he suggested the two taxa may be conspecific. Isolating mechanisms based on pheromones remain unknown in anurans (Wilson, 1970). There is a slight asynchrony in the breeding seasons of the interacting taxa, but overlap is significant, and temporal isolation is not apparent during the few weeks when most eggs are laid. The nature of this asynchrony is difficult to quantify (indeed
I overlooked it at the time I was working in the field, but its form is amplified if one compares data on breeding away from the hybrid zones with data from around the contacts. Near sea level more northerly populations of *P. bibroni* and *P. dendyi* breed several weeks later (Fletcher, 1889; Harrison, 1922; Frauca, 1965; Woodruff, 1972b), and more southerly populations of *P. semimarmorata* breed a few weeks earlier (English, 1910; Littlejohn and Martin, 1965, 1974) than they do in the areas of their interaction. Within the hybrid zones the asynchrony is much harder to establish because the composition of the hybrid population at a single site has never been monitored throughout a breeding season. Nevertheless, consistent trends can be seen in the available data, and in view of their possible significance in the *Pseudophryne* case, which is complicated by male territoriality and an unusual mating system, it is worth summarizing some of the evidence here.

Consider the record for the *P. semimarmorata-P. bibroni* contact near Wallan (Figure 9.2). At locality D where 44 adults were collected over six seasons, nine of ten typical *P. semimarmorata* were collected before the end of April and three of four typical *P. bibroni* were found in May. At locality E, 19 adults were collected over two seasons; typical *P. semimarmorata* and hybrids were found in equal numbers during April, whereas the single sample collected in May also contained typical *P. bibroni*. At locality F (46 adults, eight seasons), samples collected in April contained either no hybrids or *P. semimarmorata*-like hybrids (with hybrid indexes, HI equal to or less than three). Two samples collected later in the season both contained hybrids, and they were more *P. bibroni*-like (mean HI = 4.5, range 1–8). Finally, small samples were obtained at about the same level on the north-south transect at the western two localities designated Y. The mean HI ranged from 5.25 on March 16, 1965, to 8.66 on May 2, 1965. Although this evidence for an increase in the frequency of *P. bibroni*-like individuals as the season progresses is somewhat anecdotal, the same pattern was observed in two other areas where the two species hybridize (Woodruff, 1972b). A similar asynchrony has been detected at three transects across the *P. semimarmorata-P. dendyi* hybrid zone (Woodruff, 1972b). Details will be provided elsewhere (Woodruff, in prep.).

At present there is no evidence that the temporal asynchrony in breeding activity is any greater in the hybrid zones than in adjacent allopatry. Although this asynchrony may provide for some assortative mating at the beginning and end of the breeding season, I estimate that it cannot prevent mismating during the height of the season. I conclude that premating
isolating mechanisms between these taxa are, at best, highly ineffective, and that mating probably occurs at random among the toadlets present at a site during 80% of the breeding season.

Turning now to the evidence for postmating isolating mechanisms, I could detect no sign that the hybrids suffered sterility or ethological isolation. In vitro hybridization experiments revealed no gametic incompatibility between *P. semimarmorata* and *P. bibroni*. Fertility was 100% in a hybridization experiment between a female *P. semimarmorata* and a male *P. dendyi*, but ova in three reciprocal crosses were infertile. These last results require confirmation and are not supported by field observations, which indicate a high degree of gametic compatibility. In contrast, the evidence for hybrid inviability is good and based on both field observations and laboratory experiments. The major component of this inviability appears to be developmental incompatibility during the embryonic stage. Both natural and artificial hybrids showed higher embryonic mortality than homospecific embryos. I conclude that postmating isolating mechanisms are not sufficiently developed to prevent the occurrence of both F₁ and backcross hybrids. Yet divergence has proceeded to the point where genetic incompatibilities render the hybrids ill-adapted vehicles of gamete wastage.

Although hybrid inviability would seem to provide a basis for selection against hybridization, there is no evidence for the reinforcement of slight differences in premating behavior. [Postmating mechanisms are, of course, beyond the reach of direct selection (Muller, 1942).] All the evidence points to the conclusion that, despite interbreeding for possibly thousands of generations, little progress has been made toward perfecting intrinsic isolating mechanisms. Similarly, there is no evidence that the pairs of interacting populations are fusing, despite demonstrable gene flow in both directions. Three factors (that both parental phenotypes are maintained at the center of the zone, the great variability of the hybrid populations, and the extreme narrowness of the zone) indicate, however, that interpopulation gene flow has been reduced to extremely low levels. The frequency of hybrid phenotypes decreases very rapidly on either side of the zone, and introgression is apparently severely limited. It appears, therefore, that natural selection has been holding its own: The hybrids are ineffective as bridges for gene flow, and the interacting populations are effectively isolated from one another.

Why, then, has selection failed to perfect premating isolating mechanisms to prevent gamete wastage? Is it because of circumstances peculiar to these specific cases? For example, it may be argued that reinforcement
of slight differences in breeding season is impossible because the season is strictly circumscribed by unfavorable climatic conditions. Alternatively, perhaps the lack of a middle ear prevents these taxa from utilizing complex acoustic signals that serve as premating isolating mechanisms in many other groups of anurans. Actually, as Bigelow (1965) noted, the evolution of mechanisms that prevent interbreeding is not likely to occur in a narrow hybrid zone at all. The zones of contact involve only a very small fraction of the total range of the interacting populations. The vast majority of each species live outside the zone and have purely homospecific ancestry. Under these conditions, selection will produce mechanisms to inhibit interbreeding between the peripheral populations only incidentally, and by chance. Even if assortative mating genes do appear, and are selected for in the hybrid zone itself, their spread will be severely limited by the slope of the selection gradient, low levels of gene flow into the zone, and the fact that the hybrids suffer developmental problems (Endler, 1977). The last factor tends to keep the hybrid zone narrow as selection operates before migration.

I have concluded that these narrow hybrid zones represent the relatively stable outcome of interactions of unknown origin (Woodruff, 1979). Gamete wastage through hybridization is presumably balanced by gene flow into the zone from adjacent homospecific populations. In the absence of effective premating isolation, homospecific matings in the center of the zone may be too rare to provide a basis for the evolution of reinforcement. The interactions are interpreted, therefore, not as a race between reinforcement and fusion, but rather as a relatively stable balance between the two, in which adequate reinforcement cannot evolve, and where introgression is severely limited.

Empirical evidence to confirm this hypothesis is not yet available. Studies to determine the extent of genetic differentiation between the interacting taxa, the fitness of the various phenotypes across the hybrid zones, the extent of assortative mating, and the direction and magnitude of gene flow have not yet been conducted. In the meantime, considerable support for this interpretation comes from recent advances in population genetics (reviewed by Endler, 1977; Barton and Hewitt, Chapter 7 of this volume). Numerous theoretical treatments point to the conclusion that persistent hybrid zones will form when gene flow balances losses due to hybridization. The width of such stable zones will be related to the dispersal propensities of the interacting taxa and the relative fitness of the hybrids. In theory such zones will migrate across the country if the relative contributions from the homospecific populations are imbalanced, or if the
environment changes. These predictions all lead to numerous ideas that might be tested in nature.

McDonnell et al. (1978) have recently reported an independent analysis of the *P. semimarmorata*-*P. bibroni* interaction near Wallan. In addition to reexamining the specimens collected in the 1960s, they resampled several of the sites in the period 1972-4. Their findings on morphology and embryonic mortality tend to confirm my own. Their discovery that, in this area, *P. semimarmorata* and *P. bibroni* are fixed for alternate alleles of heart lactate dehydrogenase (*Ldh*) adds a new dimension to the story. The distribution of *Ldh* heterozygotes is quite asymmetric about the morphological hybrid zone; whereas the southern limits of the hybrid zone based on morphological, embryonic mortality, and *Ldh* criteria coincide, the northern limits based on each of these criteria differ. This is especially true for *Ldh*, which has a replacement zone about three times as wide as the morphological zone.

McDonnell et al. (1978) cite this asymmetry, coupled with an alleged change in the mean hybridity, to conclude that *P. bibroni* and the zone moved south during the period 1960-74. This point is contrary to my earlier conclusion that the zone was stable; so it deserves comment here. In addition to establishing seasonal within-locality variation, I sought evidence of annual trends in mean hybridity at four localities in this area; the data are summarized in Figure 9.5. McDonnell et al. rescoring the hybridity of all the specimens I examined from localities D, F, and G (Figure 9.2) during the period 1960-8 and compared these specimens to those they collected during 1972-4; their data are also summarized in Figure 9.5. [I have extracted these data from McDonnell et al., 1978: Figure 3, and made allowances for errors in that figure, the most significant of which involves locality D (1968), where the cumulative frequency of the animals is shown as 1.5 rather than 1.0.] Also shown in Figure 9.5 are data for 1969 from Woodruff (1972b); Woodruff (1972b) that McDonnell et al. ignored. Although there is a good correlation (+0.89) between our estimates for the mean hybridity of the 1960-8 samples, there is some disagreement on the scoring of individual specimens, and some of this could be due to the alteration of color character states following preservation. Nevertheless, as Figure 9.5 suggests, I concluded that the zone was stable during the period 1960-9 and McDonnell et al. reached a different conclusion for the period 1960-74. Unfortunately, the issue cannot be resolved in the absence of information on the collection dates for the 1972-4 samples, the more *P. bibroni*-like nature of which may simply reflect the fact that they were collected later in the breeding season.
The asymmetrical distribution of the genetic change about the morphological hybrid zone is also open to an alternate interpretation; it may be due to differential northward introgression of an \textit{Ldh} gene rather than southward movement of the zone. The imbalance may arise from the fact that \textit{P. semimarmorata} males occupy the breeding sites first and contribute more genes to the interaction. More detailed studies have in fact shown that morphology is a poor indicator of the genodynamics of hybrid zones, and that gene exchange extends into populations well beyond the zones as classically defined (Patton et al., 1979). The well-documented interaction between mice in Denmark involves a morphological zone that has not shifted in 20 years, and the asymmetry of the genetic zone in that case is clearly due to differential introgression (Hunt and Selander, 1973). Until a more critical study is undertaken, the contention that the \textit{Pseudophryne} zone near Wallan is moving south should be regarded as not proven. I conclude that the \textit{Pseudophryne} hybrid zones of southeast Australia remain problematic. Although their proximity to several universities should guarantee them continued attention, the brief breeding season and

Figure 9.5. Changes in the hybridity of sequential samples from localities D–G in the hybrid zone between \textit{P. semimarmorata} and \textit{P. bibroni} near Wallan (Figure 9.2). Range and mean hybrid index are shown for samples described by Woodruff (1972b) for the period 1960–9 (left) and McDonnell et al. (1978) for the period 1960–74 (right). See text for discussion.
small size of the adult populations will thwart the casual investigator. Nevertheless, the fact that they are now fairly well documented should allow others to make relatively more progress than I was able to in the 1960s; our questions are now more sharply defined, and techniques for the genetic characterization of these zones are now routine. With a thorough genetic analysis of these interactions, more information on the pre- and postmating isolating mechanisms involved, and some measure of the relative densities of toadlets across the zones, we should be in a better position to answer the larger questions posed by these phenomena. The fact that these interactions occur in Bassiana, a natural laboratory of considerable interest to evolutionary biologists (Darlington, 1965; Williams, 1974; Woodruff, 1973b, 1975d), should provide further impetus for the completion of this investigation.

Cerion hybrid zones in the West Indies

Among land snails, the West Indian genus Cerion is remarkable for its great display of shell variation. Although intrapopulation variation in shell size, shape, color, and sculpture is not unusual, interpopulation variation is extreme. The dramatic differences often found between adjacent populations have contributed to the recognition of over 600 “species” whose distribution appears to constitute a haphazard crazy-quilt along the coasts of Cuba and the Bahamas. With a very few exceptions the different morphotypes do not occur sympatrically. The group has attained some notoriety because two highly regarded systematists have despaired at the difficulty of applying the biological-species concept to it (Clench, 1957; Mayr, 1963, 1970). The situation is further complicated by the occurrence of numerous hybrid zones separating dissimilar morphotypes. Cerion is the classic example, in animals, of the acquisition of morphological differences without reproductive isolation.

Recently, Stephen Jay Gould and I have developed a way of freeing Cerion from its taxonomic overburden. By combining detailed mapping of geographic variation in the field with laboratory studies of anatomy, genetics, and shell morphology, we have demonstrated that the systematic problems of these highly variable organisms are not intractable (Gould et al., 1974; Woodruff, 1975a). Gould has sought to characterize the shells of each population morphometrically by taking advantage of two of Cerion’s special features. First, there is a sharp and recognizable transition between the embryonic shell (protoconch) and the accretionary shell. As the protoconch is retained in the adult shell, its terminus provides a biological criterion for the numbering of whorls and facilitates standardizing
measures at various stages of ontogeny. Second, as the shell approaches adult size, the snail changes its direction of coiling, secretes a definitive lip, and ceases growth. This fact permits the sorting of ontogenetic from other variation in the adult shell, something that is impossible in most molluscs. Taking advantage of these attributes, we may characterize any population morphometrically on the basis of the study of covariation among 19 shell variables (Gould et al., 1974). Taking a different approach, I have been able to characterize each population genetically by using standard electrophoretic procedures to survey allozymic variation in foot-muscle extracts (Woodruff, 1975b). I have now examined variation in 20–30 allozymes in about 5000 snails from 250 localities representing nearly 200 “species.” *Cerion* populations typically have moderate amounts of genetic variation: mean number of alleles per locus, 1.65–1.70; frequency of polymorphic loci per population, 0.15–0.30; and frequency of heterozygous loci per individual, 0.054–0.128. The frequencies of the different genotypes segregating at polymorphic loci indicate that *Cerion* are outbreeding despite their hermaphroditism.

The coordinated investigation of multivariate morphometrics and biochemical genetics of *Cerion* populations (on the same snails when possible) led to our discovery that the distribution of the various morphotypes was far from haphazard (Gould and Woodruff, 1978; Woodruff, 1978c). In the Bahamas and elsewhere, within situations that previously involved hundreds of “species,” we have begun to discern fairly simple biogeographic patterns involving relatively few morphologically variable taxa with allopatric or parapatric ranges. Although our work is far from complete, it appears that these taxa may be characterized on the basis of the different patterns of morphological covariation. The interesting thing is that similar differences are not found in studies of allozyme variation or anatomy. The morphological differences we have studied were acquired without detectable differentiation at a wide range of structural gene loci. Similarly, anatomical studies of various organ systems (including the genitalia) failed to reveal much variation among a range of morphologically distinctive “species” (Chung, 1979). It is becoming clear that the complex differences in adult shell form can be traced to relatively simple differences in developmental rates expressed during ontogeny (Gould, 1977; Galler and Gould, 1979). It is quite possible that all of *Cerion*’s diversity may ultimately be under the control of a few regulatory genes. The genetic uniformity was a source of some embarrassment; we had hoped that a molecular technique might succeed in revealing phylogenetic relationships where traditional methods had failed. It was not until we
turned our attention to the hybrid zones between the various morphotypes that the approach paid off; the hybrid zones are typically areas of genetic anomaly. The frequency of these zones in Cuba and the Bahamas (where we have found more than 20) suggests that we will not understand Cerion until we understand the history and significance of these hybrid zones.

Five hybrid zones will now be described to illustrate the range of phenomena we have encountered. Detailed reports of these and other interactions will appear elsewhere in due course.

Hybridization between \textit{C. abacoense} and \textit{C. bendalli} near Rocky Point, Great Abaco, Bahamas

This interaction involves a ribby morphotype (\textit{C. abacoense}) and a mottled morphotype (\textit{C. bendalli}) and has been described in broad outline by Gould and Woodruff (1978). The original analysis was based on over 400 specimens from 11 localities; our conclusions are supported by the subsequent study of an additional 255 snails. A zone of allopatric hybridization about 500 meters wide separates the parental populations. The snails are continuously and abundantly distributed along the linear transect close to the shore. Morphologically, the geographically intermediate populations display a gradual and continuous transition in mean phenotype. There is no increase in morphometric variability in the hybrid samples. Genetically, the parental species are almost identical (Nei’s $I = 0.98$); near Rocky Point 15 of the 20 allozymes studied are monomorphic and fixed for the same allele in each taxon. Variation was detected at five loci: malate dehydrogenase ($Mdh-1$), leucine aminopeptidase ($Lap-1$), glutamic oxalacetic transaminase ($Got-1$), 6-phosphogluconate dehydrogenase ($6-Pgdh-1$), and a nonspecific esterase ($Est-2$). In the case of $Mdh-1$, $Lap-1$, and $Got-1$ there was no significant change in allele frequency across the hybrid zone. In addition, there was no significant change in the proportion of heterozygotes of these allozymes in the hybrids. In contrast, both $6-Pgdh-1$ and $Est-2$ show significant changes in allele frequency associated with the hybrid zone (Figure 9.6). In the case of $6-Pgdh-1$, an unexpected allele, $6-Pgdh-1'$, was found at high frequency in populations from within the morphological hybrid zone and at decreasing frequency over a distance of 2.5 km to the south. Similarly, in $Est-2$ the common allele in both parental species, $Est-2^c$, was replaced by a typically rare allele, $Est-2^d$, over much the same area. In addition, two alleles that were very rare elsewhere, $Est-2^a$ and $Est-2^b$, occurred at low but significant frequencies in the genetic hybrid zone.
Hybridization between *C. glans* and *C. gubernatoria* on New Providence, Bahamas

A second interaction between ribby (*C. glans*) and mottled (*C. gubernatoria*) morphotypes is being studied on New Providence. Despite the phenotypic and ecological similarities to the previous species pair, the snails on New Providence are anatomically distinct from their counterparts on Abaco (Chung, 1979). The hybrid zone between these morphotypes is also different in several respects from that at Rocky Point. North of the Nassau International Airport we found the hybrids displayed a host of unusual phenotypes and considerable variation in shell size. The hybrids are continuously distributed between the parental types, and the allopatric hybrid zone appears to be less than 500 meters wide. The snails are actually more abundant in the hybrid zone than elsewhere, but this phenomenon may be the result of extensive recent land clearing in the area. Genetically, the hybrids show greater variability than their parental populations, which, as on Abaco, are indistinguishable from one another at the loci surveyed. Morphologically intermediate samples show higher levels of heterozygosity in *Mdh-1* and *Est-2*; in the latter enzyme a fast

![Figure 9.6. Allozyme variation associated with the hybrid zone (shaded) between *C. abacoense* (left) and *C. bendalli* (right) near Rocky Point, Abaco. Changes in allele frequency are shown for 6-phosphogluconate dehydrogenase-1* (upper) and esterase-2*, -2, and -2* (indicated by a circle) (lower). The morphological hybrid zone is about 500 meters wide.](image)
electromorph unique to the hybrid zone rises to a frequency of 0.10. [An earlier statement (Woodruff, 1978c) that this hybrid zone was not an area of genetic anomaly now seems to have been premature.] Whether this unexpected allele is asymmetrically distributed across the hybrid zone and whether the zone of increased genetic variability is wider than the morphological zone are not yet clear.

Figure 9.7. Map of Long Island, Bahamas, showing the position of some of the hybrid zones between dissimilar morphospecies of Cerion. Ranges identified are for, from left to right, *C. malonei*, *C. coeruleus*, *C. stevensoni*, and *C. ferdandina*. Position of the hybrid zone illustrated in Figure 9.8 is indicated by the large arrow.
Hybridization between C. stevensoni and C. fernandina on Long Island, Bahamas

On Long Island we have located 12 hybrid zones between the seven distinct morphotypes we recognize (Figure 9.7). Spectacular transitions involve the large white, smooth-shelled C. fernandina, the squat, white, smooth-shelled C. malonei, and the distinctive member of the subgenus Umbonis, C. stevensoni. Defined on the basis of shell morphology, these hybrid zones are typically less than 500 meters wide, and some are only 100 meters wide. Whereas some zones are associated with changes in snail abundance or habitat, others are not. Although the various species involved are very similar to one another genetically, I have again found genetic anomalies associated with all of the hybrid zones examined. The hybrid populations show significantly greater variation in allele frequencies and are characterized by novel genotypes absent in adjacent parental populations. The allopatric hybrid zone between C. stevensoni and C. fernandina is now reasonably well documented and shows these effects in Mdh-1, 6-Pgdh-1, and Est-2 (Figure 9.8). Furthermore, as at Rocky Point, the area of genetic anomaly is three to four times as wide as the morphological transition and asymmetrically distributed about it; the

Figure 9.8. Allozyme variation associated with the hybrid zone (shaded) between C. stevensoni (left) and C. fernandina (right) on Long Island. Changes in allele frequency are shown for 6-phosphogluconate dehydrogenase -1", -1", and -1" (upper) and malate dehydrogenase -1", -1", and -1" (indicated by a circle) (lower). The morphological hybrid zone is about 300 meters wide.
A genetic effect was detected abruptly at the *C. stevensoni* end of the morphological hybrid zone but extends about 600 meters into the range of *C. fernandina*.

**Hybridization between *C. moralesi* and *C. geophilus* on the Banes Peninsula, Cuba**

The most widely publicized *Cerion* hybrid zones involve a series discovered by Ernst Mayr in the 1950s, but known only from his map and a single interpretive paragraph (Mayr, 1963:399, 1969, 1970). Recently, Galler and Gould (1979) have published a detailed report on the interaction between *C. moralesi* and *C. geophilus* based on the shells Mayr collected. *C. moralesi* has a smooth, mottled shell; *C. geophilus* has a squat, ribby shell and a spiral sculpture of numerous incised lines that pass over the axial costae. This latter feature is a characteristic of the subgenus *Umbonis* (Clench and Aguayo, 1952). Galler and Gould studied the shell morphology of Mayr’s four samples and concluded that the zone of allopatric hybridization was less than 600 meters wide. Mayr’s field notes indicated that the snails were abundant and continuously distributed across the hybrid zone. Galler and Gould found no evidence for increased variability in the morphologically and geographically intermediate samples. They demonstrated the existence of several univariate clines and one multivariate cline covering the zone. They concluded that the large morphological differences between the contrasting morphotypes stem from a small alteration in the rate of shell widening during the early postembryonic phase of growth. Nothing is known about the genetics of this interaction.

**Hybridization between several morphotypes in the Bimini Islands, Bahamas**

Mayr and Rosen (1956) found pronounced but geographically irregular morphological variation among the *Cerion* of six small islands in the Bimini group. They interpreted the pattern to be the result of widespread hybridization between three essentially allopatric and variable morphospecies. Hybrid colonies were shown to be more variable than homospecific ones with respect to sculpture and color, but no such effect was detected for size or shape. Unfortunately, the geographic complexity of this situation precludes a simple discussion of their results along the lines developed in the foregoing accounts. White (1978), for example, has pointed out that one of the highly variable “hybrid” colonies is actually located between two very uniform colonies, which are very similar to one
A reanalysis of this situation using multivariate and genetic techniques is now in order.

Although the uneven documentation prevents a rigorous comparison of these interactions, they all appear to involve narrow zones of allopatric hybridization (sensu Woodruff, 1973a). They may, or may not, be associated with changes in snail density, or genetic and morphometric variability. In two well-studied cases, the morphological hybrid zone is narrow and lies at one edge of a broader zone of genetic anomaly. Although some of the hybrid zones are associated geographically with environmental changes, others apparently are not. It must be emphasized that no single explanation should be sought to interpret such a range of phenomena; the zones may be of diverse origin and significance.

Mayr concluded that the hybrid zones he studied on Cuba and Bimini were the result of secondary contacts between forms that had differentiated in isolation. The checkerboard distribution pattern of the various morphotypes was presumably the result of infrequent long-distance dispersal by hurricanes. In contrast, our own work suggests that some of the dissimilar populations may have arisen in situ as a result of parapatric differentiation (Woodruff, 1978c). Rather than the checkerboard distribution pattern perceived by earlier workers (results that were partly a consequence of taxonomic malpractice), we find fairly regular patterns of geographic variation over large areas of the Little Bahama Bank (Gould and Woodruff, 1978), the Great Bahama Bank (Woodruff, 1978c), Great Inagua (Gould and Woodruff, in prep.), the Greater Antilles (from Hispaniola to the Virgin Islands; Gould and Paull, 1977), and the Dutch Leeward Islands (Gould, 1969). Wherever we have looked, we have found that forms characterized by the most divergent shell types share a common internal anatomy and set of allozymes. The finding that some major differences in shell morphology arise as a result of relatively simple heterochronous changes in ontogeny also removes one of the objections that earlier workers may have raised to the "role of parapatric differentiation in Cerion. Perhaps we will find Cerion to be capable of very close evolutionary "tracking" of microenvironmental heterogeneity; the analogy with some variable plants, in which highly localized, specially adapted ecotypes replace one another over distances of a few meters, is striking (Bradshaw, 1972). Unfortunately, present techniques do not allow us to discriminate between primary and secondary zones of hybridization. The Cerion case is interesting in that it probably contains both types; the challenge is to learn how to distinguish them.
If the diverse origin of these hybrid zones is presently shrouded in mystery, can anything predictive be said of their future? Are they stable or transient phenomena? (And if transient, will the adjacent populations fuse or interact so that some isolating mechanism is reinforced and speciation completed?) *Cerion*’s fossil record is of little use beyond establishing that some morphotypes have occupied parts of their present ranges for thousands of years. (We have yet to find a fossil hybrid zone.) Sequential sampling of snails across a hybrid zone is also unlikely to resolve the issues quickly because generation time is relatively long (probably 5 years; Woodruff, 1978c) and sample sites must be located with much greater precision than has been customary in the past. We are also profoundly ignorant of the antihybridization mechanisms that *Cerion* may employ. Statements to the effect that reproductive isolating mechanisms are not easily acquired in this genus are quite plausible but not substantiated. Thus, although our morphological and genetic data indicate that hybrid zones on Abaco and Long Island involve fully compatible morphospecies, the zones are very narrow. There is no evidence for assortative mating, and there is no evidence that fusion is occurring. Although the genodynamics of these interactions remains obscure, it appears that some zones may be relatively stable.

The behavior of the *Cerion* morphospecies would seem to be in clear violation of the biological species concept. Actually, the problem may not be with *Cerion* but with a species concept that overemphasized the importance of reproductive isolation. It is becoming clear that genetic isolation cannot be directly equated with reproductive isolation. Common homeostatic mechanisms and gene flow may protect the integrity of the gene pool even in the absence of reproductive isolation (Bigelow, 1965; Hunt and Selander, 1973; Endler, 1977; Woodruff, 1979). If one allows that semispecies may hybridize without losing their integrity, then these *Cerion* are just extremely variable examples of biological species. The fact that morphologically different populations share a common set of structural genes has no bearing on the speciation question; there is little evidence for the extensive reorganization of gene pools during speciation (Throckmorton, 1977; Nevo and Cleve, 1978). Nevertheless, some genetic differentiation has occurred, as evidenced by the changes in the patterns of covariation among morphometric traits and the genetic anomalies associated with the hybrid zones themselves. The latter phenomena have recently been reported for hybrid zones involving semispecies of *Mus* (Hunt and Selander, 1973) and *Rana* (Sage and Selander, 1979); and Selander and I (in prep.) have discussed the roles of mutation and intragenic recombination in producing these effects.
I do not think that the interpretive difficulties I have encountered with the *Pseudophryne* or *Cerion* hybrid zones are unique. They are merely a reflection of our present scientific inadequacies; specifically, we lack appropriate techniques to test the various alternate hypotheses concerning the origin and future of these phenomena. Progress in this area would appear to require the development of field methods for the characterization of the genodynamics of each situation. If we can monitor the net movement of genes into and across a hybrid zone today, then we should be able to simulate the behavior of that zone at other points in time. I am pursuing this idea with *Cerion*, an organism in which dispersion and dispersal can be estimated both accurately and easily, and I have been conducting pilot studies in populations adjacent to the Rocky Point hybrid zone since 1973. The purpose of these experiments, which involve monitoring the movements of large numbers of snails, is to determine the feasibility of estimating the gene flow parameter. [Gene flow is typically defined as the product of the mean distance traveled in a generation and the square root of probability of leaving a deme or neighborhood (May et al., 1975).] Preliminary results indicate that mean dispersal at a site can be estimated relatively quickly because annual displacements for both juveniles and adults are very similar, and successive annual displacements have not varied appreciably during 5 years; the dispersal estimate for adults for 1 year is nearly the same as the estimate based on a whole generation. Displacements of individual snails at one site are shown for two successive 6-month periods in Figure 9.9. At the two sites monitored it is already apparent that although mean displacement is very small (less than 3 meters), and maximum detected displacement is about 20 meters, gene flow is 50% higher at the northern than at the southern end of the hybrid zone. It is hoped that studies of this type will not only contribute to the explanation of the asymmetrical aspects of some hybrid zones but also lead to a better understanding of their basic genodynamics.

In my opinion one of Michael White's major contributions to evolutionary biology has been the demonstration that the classical sympatric and allopatric models of speciation no longer satisfactorily account for the observed diversity of animals. By very careful attention to cytogenetic details he has helped bring about a change in the way we think about the processes of speciation. With the steady diminution of the alleged role of "cohesive forces" in preventing speciation (Ehrlich and Raven, 1969; Lewontin, 1974; Felsenstein, 1975; Levin, 1979), our focus has shifted to the origin of genetic isolating mechanisms rather than the geographic subdivision of the gene pool as the prime cause of speciation. White's work on the morabine grasshoppers illustrates this well and constitutes one of
small number of examples that set the stage for the ongoing revitalization of the field. I think it is a tribute to the generality of his vision that the processes he elucidated in the morabines are also relevant to the evolution of the *Pseudophryne* and *Cerion*, two groups in which chromosomal rearrangements have not been detected. His continued insistence that speciation represents far more than "a category of biogeographic acci-

Figure 9.9. Dispersal of individual cernion at a study site near Rocky Point, Abaco. The displacement of individually marked snails initially found in a 55 m² area is shown for the period November 1974 to May 1975 (upper) and May 1975 to November 1975 (lower). During the former period 227 snails moved, whereas 184 moved during the latter. Snails that did not move during each period (59 during the winter, 32 in the summer) are not shown. Axes show distances in centimeters.
dents" (White, 1959) has helped to bring this crucial evolutionary process back into the mainstream of biological inquiry.

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References


David Woodruff


Genodynamics of Hybrid Zones
