

MAINTENANCE OF A NARROW HYBRID ZONE IN *GEOMYS*: RESULTS FROM CONTIGUOUS CLUSTERING ANALYSIS

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ABSTRACT.— Contiguous clustering was used to analyze the distributions of parental, hybrid, and backcross individuals within a contact zone between *Geomys bursarius major* and *G. knoxjonesi*. Clustering was performed using the characteristics of three electrophoretic loci, mtDNA, diploid number, and proportion of the genome represented by *G. b. major*. Results showed that significant contiguous groups were comprised predominantly of either parental type or hybrid and backcross individuals. Clusters derived from the different genetic markers were significantly correlated and thus gave similar results for structure of the hybrid zone. Significant contiguous clusters of hybrid individuals (except for mtDNA where identification of hybrids was not possible) were confined to intermediate areas between large clusters of each parental type. The presence of clusters clearly identifiable as parental and backcross groupings together with significant linkage disequilibrium values between most loci pairs and frequency distributions for genetic combinations support the dynamic equilibrium model for maintenance of this hybrid zone between the two *Geomys* species. Results for this hybrid zone were somewhat different from those evident for a zone of intergradation between two chromosomal races of *Peromyscus leucopus*. Similar analyses showed that *P. leucopus* had no clusters of parental types and the hybrid equilibrium model was most compatible with hypotheses regarding maintenance of that zone. The contiguous clustering method, together with other more conventional techniques, has proven to be of considerable utility for the assessment of the dynamics of hybrid zones.

Key words: *Geomys*, *Peromyscus*, hybrid zones, contiguous clustering, linkage-disequilibrium

Because hybrid zones represent a unique situation in which relative fitness of individuals, intermediate stages of reproductive isolation, gene dynamics, and the molecular basis of new genetic variation can be examined in natural populations, hybrid zones have been of considerable interest to evolutionary biologists (Baker et al., 1989; Simmons et al., 1992; Bradley et al., 1991a, 1993). Providing a synthesis of hybrid zone biology has been difficult because 1) of the many alternative perspectives from which hybrid zones can be viewed and studied, 2) information from one hybrid zone has proven to be a poor predictor of the characteristics of other hybrid zones, and 3) forces involved in the maintenance of hybrid zones are complex (ecological to genetic), synergistic, and difficult to factor into independent components.

Several papers have addressed statistical procedures that provide insight into the biological forces that create and maintain such narrow hybrid zones (Barton and Hewitt, 1981, 1985, 1989; Endler, 1977; Kaneshiro, 1976, 1980; Moore, 1977; Shaw, 1981; Templeton, 1981; Woodruff, 1981; Van Den Bussche et al., 1993.). Van

Den Bussche et al. (1993) have outlined the predictions of gene distributions for three alternative models proposed to maintain hybrid zones (Table 1). To better understand how statistical descriptors of hybrid zones vary, we have attempted to collect comparable data sets from two disparate zones. One of these two zones has been described elsewhere (Van Den Bussche et al., 1993). Of the three alternative models, the most probable explanation for the maintenance of the *Peromyscus leucopus* hybrid zone is the hybrid-equilibrium model. Linkage disequilibrium statistics indicate a significant association between two of the five diagnostic markers; the zone is characterized by many small clusters, and only two markers show concordance of clusters (chromosomes 5 and 20) suggesting that the progeny are the result of interhybrid matings; and finally, frequency histograms of the genotypic data are compatible with the conclusion that most offspring are the result of interhybrid crosses and that hybrid individuals are encountering little or no selection, either positive or negative, relative to the parentals (hybrid-equilibrium; Endler, 1977).

Table 1. Conditions of expression of linkage equilibrium, contiguous clustering, and genotypic class structure that support either the dynamic-equilibrium, hybrid superiority, or hybrid equilibrium models for a population in the center of a hybrid zone as proposed by Van Den Bussche et al. (1993).

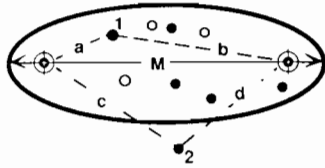
Model	Linkage disequilibrium	Contiguous clustering	Genotypic classes
Dynamic-equilibrium	Significant linkage disequilibrium due to a paucity of alternative homozygotes for genotypic classes expected from backcrossing.	If clusters exist, they will preferentially consist of pure parental and F1-types. Structure correlated.	a) Excess of parental genotypes. b) Excess of F1-types. c) Alternative homozygotes less than expected.
Hybrid-superiority	a) Lack of significant linkage disequilibrium. b) In cases of strong selection favoring hybrids, paucity of pure parental types.	a) No structure, or few significant clusters. b) Clusters consist of hybrids, and structure of genetic characters are not correlated.	Hardy-weinberg expectations or paucity of parental types.
Hybrid-equilibrium	Significant linkage disequilibrium not in paucity of parental types.	Structure present for two or more genetic characters, and the patterns of pairwise comparisons is correlated. Clusters not due to parental types.	Hardy-Weinberg expectations of paucity of parental types.

The second hybrid zone involves pocket gophers of the genus *Geomys* (Baker et al., 1989; Bradley et al., 1991a,b). Van Den Bussche et al. (1993) proposes three methods for determination of the forces responsible for the maintenance of hybrid zones: 1) frequency of genotypic classes; 2) linkage disequilibrium analysis, and; 3) contiguous clustering analysis. The results of a single analysis will not differentiate between the three models due to the various factors affecting the gene distributions in hybrid zones. However, with the proposed suite of analyses, it is possible to eliminate some of the alternatives. Although all three tests have not been performed previously on the *Geomys* contact zone, frequency histograms of the genotypic data suggest that the *Geomys* contact zone is concordant with the expectations for a zone that is maintained by strong selection against hybrids (dynamic-equilibrium, Moore, 1977; Baker et al., 1989). Our objective in this manuscript is to complete the linkage disequilibrium and contiguous clustering (Chesser and Van Den Bussche, 1988) analyses to more fully understand the evolutionary forces responsible for maintenance of the hybrid zone involving *Geomys hirsarius major* and *Geomys knoxjonesi* and to compare them with those found for *Peromyscus leucopus*.

MATERIALS AND METHODS

Seventy-five individuals were collected during the summer of 1986 and 37 additional individuals were collected during the summer of 1988 from a naturally occurring hybrid zone along a 6-km north-south transect parallel to the DeBaca-Roosevelt county line, 15.5 miles S and 3.0 miles E of Taiban, New Mexico (Baker et al., 1989; Bradley et al., 1991b). See Pembleton and Baker (1978), Baker et al. (1989), and Bradley et al. (1991a,b) for a description and characterization of this hybrid zone. A total of 110 of the 112 individuals previously reported in Bradley et al. (1991a) were used in present analyses (the two additional individuals were progeny of one of the females sampled). Each animal was examined for chromosome number, ribosomal DNA, mitochondrial DNA, and three protein systems (lactate dehydrogenase, Ldh-1; alcohol dehydrogenase, Adh; peptidase-B, Pep-B-1). Animals were karyotyped using the yeast-stress in vivo method of Lee and Elder (1980). Samples of liver, heart, kidney, and muscle were frozen in liquid nitrogen and stored at -80°C prior to further analysis. Voucher specimens were prepared as skins and skeletons and deposited in The Museum, Texas Tech University. MtDNA and rDNA were visualized from Southern blots (Southern, 1975) of total cellular DNA. DNA isolation from skeletal muscle followed Hillis and Davis (1986). Elec-

Criterion for Contiguity



M = length of major axis
 D = distance between foci individuals
 D_{max} = maximum distance between all individuals sampled

$$M = 2\sqrt{\left[\frac{D}{2}\right]^2 + \left[\frac{D^2}{2D_{max}}\right]^2}$$

If the sum of the distances between a point and the two foci is less than M , then that point is considered to be contiguous because it falls within the ellipse. Individual 1 is contiguous because $M > a + b$, yet individual 2 is not because $M < c + d$.

Fig. 1. Graphical description of the criterion for contiguity used in the contiguous clustering algorithm (Chesser and Van Den Bussche, 1988). The two individuals which are the foci of the ellipse are shown as crosshairs on the major axis (M). Individuals are considered to be contiguous if they fall within an ellipse, the eccentricity of which is determined by the distance between the two foci relative to the maximum distance separating samples in the population. As shown in this graph, however, the eccentricity need not be directly determined.

trophoresis, Southern transfer, and hybridization were described by Hillis and Davis (1986) and Davis (1986). For protein electrophoresis, heart kidney and liver tissue extract preparations and electrophoretic techniques followed Selander et al. (1971). Protein-staining techniques were as described in Selander et al. (1971) and Harris and Hopkinson (1977). Detailed descriptions of methods for preparing, identifying, and scoring the above mentioned systems are listed in Baker et al. (1989) and Bradley et al. (1991a,b). Linkage disequilibrium was evaluated for each pair of diagnostic loci and chromosomal types using the methods of Hill (1974).

Contiguous clustering is a method that identifies groups of spatially adjacent observations in which individuals possess a greater than expected number of a discrete character (genotype in this instance). Contiguous clustering is well suited for the analysis of genotype distributions within hybrid zones. Whereas the method does not necessarily identify specific breeding groups, it specifies the location of groups of individuals which possess identical genotypes. Thus, for hybrid zones it may identify geographic areas comprised of parentals, hy-

brids, or backcross individuals. Lack of significant clusters may indicate random mixtures of the various types (Van Den Bussche et al., 1993).

Observations are considered to be contiguous if they fall on or within an ellipse of which the foci are two individuals possessing identical genotypes for a locus (Fig 1). The eccentricity of the ellipse is determined by the distance between the foci relative to the maximum distance between any pair of observations. Thus, foci which are proximal will determine a very flat ellipse, and encompass only nearby individuals, whereas maximally separated foci will determine a circle which encompasses all observations.

Contiguous clustering was performed for all genetic characters listed in Table 2 with the exception of rDNA, which is a continuous character, in accordance with the methods described by Chesser and Van Den Bussche (1988), with some modifications. The cross entropy value (H) is determined as

$$H = \frac{a-2}{n-2} \ln \left[\frac{a-2}{(n-2)p} \right] + \frac{b-2}{n-2} \ln \left[\frac{b-2}{(n-2)(1-p)} \right]$$

where

a = the number of individuals within a potential cluster that possess the designated genotype
 b = the number of individuals within a potential cluster that do not possess the designated genotype.

n = the total number of individuals in the potential cluster, and

p = the adjusted proportion of individuals in the total population that possess the designated genotype.

$$p = \frac{\text{no. individuals with genotype}}{\text{total no. individuals}}$$

A value of two is subtracted from many of the parameters because of the nonrandom manner in which the potential clusters are initiated. Two identical genotypes are selected to initiate the clustering process (see Chesser and Van Den Bussche, 1988). The cross entropy chi square value presented by Chesser and Van Den Bussche (1988, p. 4) is appropriate for a two-tailed test. Because we are interested only in groups that have a greater number than that expected by random distribution, the Chi square value is calculated as $\chi^2 = nH$ (rather than $2nH$) and has one degree of freedom. Only groups in which $\frac{a-2}{n-2} < p$ were candidates for significant clusters. Clusters were considered significantly different from random aggregates only if the probability of the Chi square was ≤ 0.01

Table 2. Sex, chromosome number, genotypes, overall classification (Type), percent of genome classified as *G. b. major*, and spatial coordinates (x and y) representing parental *G. b. major* (M), hybrids and backcrosses (H), and parental *G. knoxjonesi* (K). rDNA values are expressed as percentage of *G. b. major*.

SEX	2N	mtDNA	rDNA	Adh	Ldh	Pep	Type	percent major	X	Y
M	72	M	100	M	M	M	M	8/8	16	20
F	70	K	0	K	K	K	K	0/8	21	6
F	72	K	85.3	H	M	M	H	7/8	15	15
F	70	K	0	K	K	K	K	0/8	15	17
F	70	K	0	K	K	K	K	0/8	19	11
F	70	M	0	H	K	K	H	1/8	19	9
F	70	K	0	K	K	K	K	0/8	19	10
F	72	M	100	M	M	M	M	8/8	16	22
F	71	K	100	H	H	H	H	4/8	17	26
F	72	M	100	M	M	M	M	8/8	16	17
F	71	K	66.4	H	H	H	H	4/8	18	21
F	71	K	55.3	H	H	H	H	4/8	16	15
F	72	M	100	M	M	M	M	8/8	14	25
F	72	K	80.0	H	H	H	H	5/8	16	24
M	71	M	75.5	H	H	H	H	4/8	15	26
F	72	M	100	M	M	M	M	8/8	17	20
M	72	M	100	M	M	M	M	8/8	19	12
M	71	K	47.1	H	H	H	H	4/8	18	12
M	70	K	5.6	K	K	K	H	0/8	16	16
F	70	K	0	K	K	K	K	0/8	20	12
F	71	K	38.7	H	H	K	H	3/8	17	16
F	70	K	0	K	K	K	K	0/8	20	19
M	72	M	100	M	M	M	M	8/8	14	31
M	72	K	97.5	M	M	H	H	7/8	15	27
M	71	K	81.2	H	H	M	H	5/8	18	15
F	70	K	13.2	K	K	K	H	0/8	17	11
F	70	K	0	K	K	K	K	0/8	19	8
M	70	K	11.7	K	K	K	H	0/8	20	10
F	71	M	52.6	K	H	H	H	3/8	17	12
M	71	K	65.0	H	H	H	H	4/8	17	9
M	72	M	100	M	M	M	M	8/8	24	10
M	71	K	35.9	H	H	H	H	4/8	18	9
F	71	K	77.9	H	H	H	H	4/8	17	31
F	72	M	100	M	M	M	M	8/8	15	19
F	70	K	52.1	H	H	K	H	2/8	10	9
M	72	M	100	M	M	M	M	8/8	14	27
F	72	M	100	M	M	M	M	8/8	18	30
M	70	K	0	K	K	K	K	0/8	18	8
M	70	K	0	K	K	K	K	0/8	15	11
M	71	K	61.9	H	H	H	H	4/8	13	28
F	70	K	0	K	K	K	K	0/8	18	10
F	71	K	59.2	H	H	H	H	4/8	17	32
F	72	M	100	M	M	M	M	8/8	14	33
M	72	M	100	M	M	M	M	8/8	15	23
F	71	K	8.3	K	K	K	H	1/8	21	10
F	71	K	52.3	H	H	H	H	4/8	19	14
F	72	M	100	M	M	M	M	8/8	15	32
M	71	K	45.4	H	H	H	H	4/8	20	14
M	72	M	100	M	M	H	H	7/8	14	30
F	72	K	62.7	H	H	H	H	5/8	16	11
M	71	K	65.3	H	H	H	H	4/8	17	13
M	72	M	100	M	M	M	M	8/8	20	15
F	70	K	0	K	K	H	H	1/8	15	4
M	71	K	51.7	H	H	H	H	4/8	15	21

Table 2. Continued

SEX	2N	mtDNA	rDNA	Adh	Ldh	Pep	Type	percent major	X	Y
F	72	M	100	M	M	M	M	8/8	13	34
F	72	K	100	M	H	H	H	6/8	1	29
M	71	K	34.6	H	H	H	H	4/8	14	11
M	70	K	0	K	K	K	K	0/8	12	3
M	71	K	0	H	H	H	H	4/8	17	2
M	72	M	88.0	M	H	M	H	7/8	18	11
M	72	M	100	M	M	M	M	8/8	1	28
F	71	K	77.5	H	H	K	H	3/8	15	29
M	72	K	100	M	M	M	H	8/8	20	29
M	72	M	100	M	M	M	M	8/8	19	44
F	70	K	0	K	K	K	K	0/8	4	7
M	70	K	0	K	K	K	K	0/8	2	5
F	71	M	74.0	H	H	H	H	4/8	11	29
M	72	M	100	H	M	M	H	7/8	17	48
M	70	K	0	H	K	K	H	1/8	2	7
M	70	K	0	H	K	K	H	1/8	6	6
M	72	M	100	M	M	M	M	8/8	15	42
M	72	M	100	M	M	K	H	6/8	19	45
F	70	K	0	K	K	K	K	0/8	3	7
M	70	K	0	K	K	K	K	0/8	17	11
F	70	K	0	K	H	K	H	1/8	18	12
F	70	K	0	K	K	K	K	0/8	15	12
F	71	K	90.6	H	H	H	H	4/8	18	12
F	71	K	26.1	H	H	H	H	4/8	18	12
M	71	M	22.9	H	H	M	H	5/8	18	12
M	70	K	0	K	K	K	K	0/8	17	12
F	71	M	100	M	H	M	H	6/8	18	12
M	70	K	0	K	K	K	K	0/8	17	28
F	70	K	0	K	K	K	K	0/8	16	38
F	72	M	100	M	M	M	M	8/8	1	18
F	72	M	97.8	M	M	M	H	8/8	16	42
M	72	M	100	M	M	M	M	8/8	19	11
-	-	K	91.3	M	M	M	H	6/6	18	12
-	-	K	76.5	-	M	H	H	3/4	18	12
F	70	K	0	K	K	K	K	0/8	3	7
F	71	K	0	K	K	K	H	1/8	20	9
M	71	M	100	H	-	M	H	4/6	34	28
F	70	K	0	K	K	K	K	0/8	17	11
F	72	K	100	K	H	K	H	3/8	24	28
M	70	K	0	K	K	K	K	0/8	18	11
F	71	M	79.2	H	H	H	H	4/8	18	12
F	71	K	92.2	H	H	H	H	4/8	18	12
F	70	K	0	K	K	K	K	0/8	16	12
F	72	M	97.1	M	M	H	H	7/8	16	28
M	72	M	100	M	M	H	H	7/8	18	12
F	72	M	100	M	M	M	M	8/8	16	19
M	72	M	100	M	M	M	M	8/8	16	25
M	72	M	100	M	M	M	M	8/8	22	6
M	70	K	0	K	K	K	K	0/8	17	11
M	71	M	100	H	H	M	H	5/8	16	29
F	70	K	0	K	K	K	K	0/8	18	6
F	70	K	93.8	H	H	H	H	3/8	18	12
F	72	M	100	M	M	M	M	8/8	16	25
F	72	M	100	M	M	M	M	8/8	16	35
F	72	M	100	H	M	M	H	7/8	16	16
F	72	M	100	M	M	M	M	8/8	16	23

Connectivity matrices with the probability of each pair of individuals being included in the same cluster (Scribner and Chesser, 1993) were output from the contiguous clustering analysis. Tests of conformity of these matrices were performed using Mantel (1967) matrix regression to determine if the clustering for each genetic character gave similar results.

RESULTS

Chromosomal, allozymic, rDNA, mtDNA, and capture coordinates for each individual are presented in Table 2. There is a general trend for a south to north gradient of parental *G. knoxjonesi*, hybrids, and parental *G. b. major* phenotypes as evidenced by rDNA (Fig. 2). East-west gradients were not evident. The overlap of hybrid, backcross, and parental types make the identification of discrete population boundaries difficult. Contiguous clustering analysis did, however, result in several significant population units.

A total of 31 unique contiguous clusters ($P \leq 0.01$) was found within this population of *Geomys b. major* and *Geomys knoxjonesi* (hereafter referred to as *major* and *knoxjonesi*, respectively; Fig 3-5). Five of these were detected for the Adh locus in which there are *knoxjonesi*, hybrid, and *major* clusters. These five clusters consisted of a single cluster containing 48 individuals primarily composed of *major* genotypes, two clusters primarily composed of *knoxjonesi* individuals containing 50 and 23 individuals respectively, and two hybrid clusters containing 22 and 21 individuals (Fig. 3). Only five of the 110 individuals did not belong to any of the five significant Adh clusters. Four significant clusters were detected for the Ldh marker. Of these four clusters, one large cluster containing 48 individuals representing the *major* gene pool was detected, one cluster of 18 individuals representing hybrid individuals and two clusters containing 26 and seven *knoxjonesi* individuals (Fig. 3). Only a single hybrid and a single *knoxjonesi* individual was detected in more than one cluster whereas ten individuals did not belong to any of these clusters.

For the Pcp-B-1 locus, six significant clusters were detected (Fig. 4). Three small clusters were detected representing the *major* genotype, one small cluster representing hybrid individuals, and two clusters representing the *knoxjonesi* genotype. Thirty-one individuals did not belong to any of these four clusters. When individuals were clustered based on their diploid number, five significant clusters were detected (Fig. 4). One cluster con-

tained 43 individuals and represented *G. major*, 5 individuals with a diploid number of 71 formed a significant hybrid cytotype cluster whereas a second cluster containing seven individuals with a hybrid cytotype and one in-

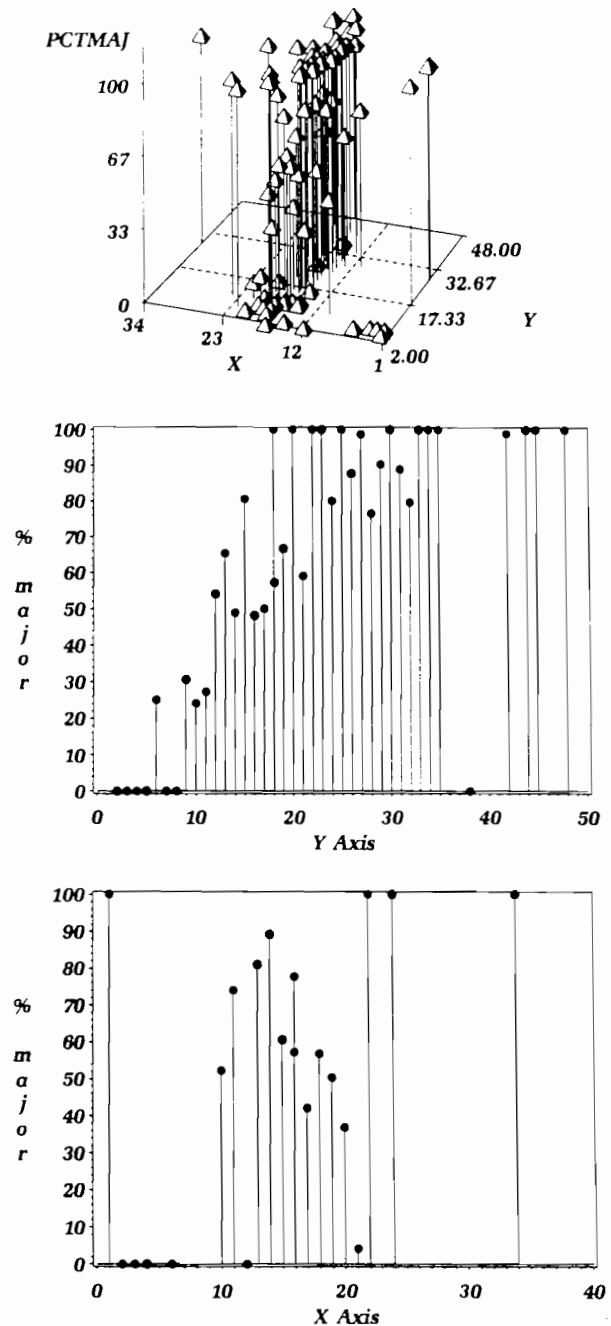


Fig. 2. Graphical representation of the distribution of individuals in accordance with their proportion of genomes being *Geomys bursarius major* (PCTMAJ) as determined from rDNA. The top figure shows the individual coordinates with the height of the sticks determined by the proportion of *G. b. major*. The bottom two figures show the average percent *major* genes for individuals along the y (North to South) and x (West to East) axes, respectively.

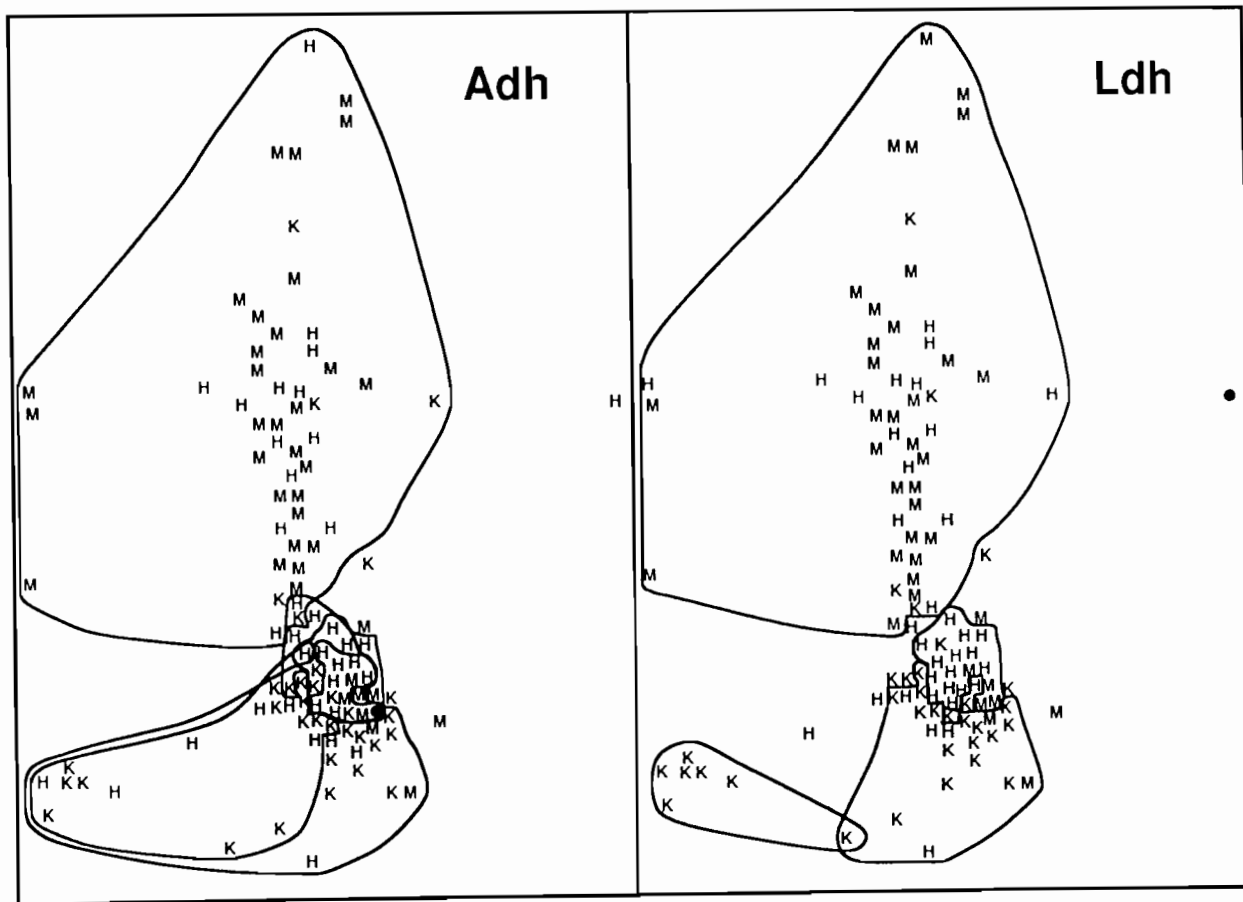


Fig. 3. Graphical representation of the position of individuals possessing specific genotypes within a hybrid zone between *Geomys bursarius major* and *G. knoxjonesi* and the results of the contiguous clustering analysis for Adh and Ldh. The solid lines represent contiguous clusters of individuals from the cross entropy X^2 ($P < 0.05$). Some distortion of the ellipses has occurred to make the figure legible.

individual with a *knoxjonesi* cytotype formed the second significant hybrid cluster. There were two significant clusters for the *knoxjonesi* cytotype (diploid number = 70) with one large cluster of 52 individuals circumscribing a smaller significant cluster of eight individuals. Only ten individuals were not included in any of the significant clusters.

When the contiguous clustering analysis was performed on mtDNA data, five significant clusters were detected (Fig. 5). Three of these clusters represented the *major* mtDNA genome and two represented the *knoxjonesi* mtDNA genome. Of the 110 individuals, only three were not included in any of the mtDNA clusters. The final clustering procedure was to classify each individual as to the relative proportion of their genome consisting of the *G. b. major* genotype. This resulted in six significant clusters, three clusters are significant due to a high number of pure *major* genomes whereas three clusters are significant due to a high number of individu-

als with pure *knoxjonesi* genomes (Fig. 5). The three *major* clusters contained 39, 38, and 14 individuals, respectively whereas the three *knoxjonesi* clusters contained 18, 11, and 10 individuals, respectively. Thirty-nine individuals were not included in any of these clusters. When all significant clusters were examined, only two individuals were not included in any of the 31 unique clusters.

Mantel (1967) analyses of connectivity matrices showed that all pairs of genetic characters were resulting in similar clustering patterns to a greater degree than would be expected by chance. Results of tests of linkage disequilibrium (Hill, 1974) showed significant linkage between ADH and LDH ($D = .053$, $P < 0.05$), ADH and chromosomal type ($D = 0.057$, $P < 0.05$), and LDH and chromosomal type ($D = 0.064$, $P < 0.05$). None of the pairwise tests of disequilibrium which included the PEP locus was significant (PEP-ADH, $D = 0.034$; PEP-LDH, $D = 0.041$; PEP-Chromosomal type, $D = 0.039$).

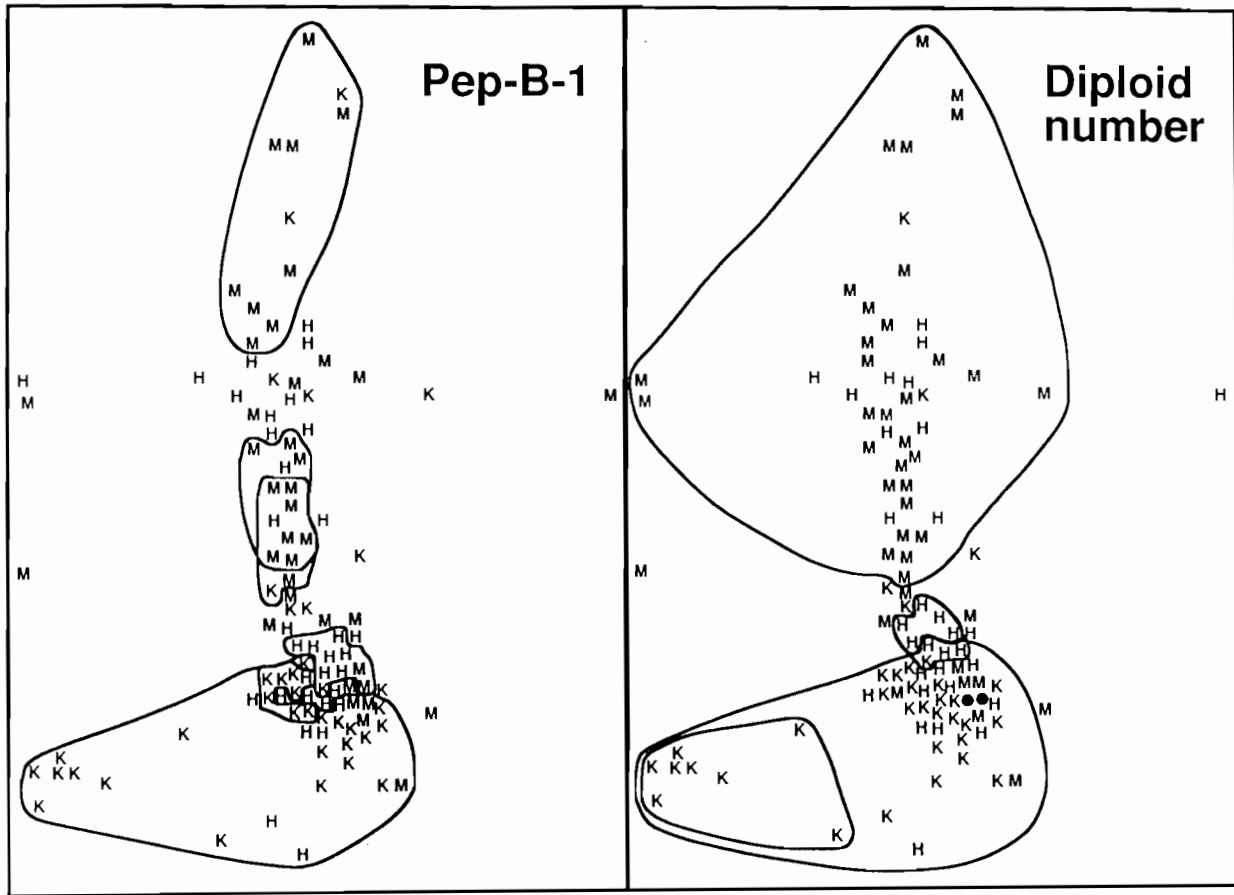


Fig. 4. Graphical representation of the position of individuals possessing specific genotypes for the Pep-B-1 locus and diploid number for individuals in a hybrid zone between *Geomys bursarius major* and *G. knoxjonesi*. Solid lines represent contiguous clusters of individuals from the cross entropy X^2 ($P < 0.05$). Some distortion of the ellipses has occurred to make the figure legible.

DISCUSSION

Van Den Busche et al. (1993) have outlined the expectations for results of the contiguous clustering analysis for hybrid zones that are maintained by the dynamic-equilibrium model. These expectations are: if clusters exist, they will preferentially consist of three types, each parental type and F₁'s and the spatial distribution patterns for individuals possessing various genotypic combinations are correlated. The stipulation that hybrids only consist of F₁ individuals is probably too stringent if they have even low fertility. With this stipulation, the results of the contiguous cluster analysis are exactly as described by Van Den Busche et al. (1993) for the dynamic equilibrium model (Table 1).

Evidence from the predictions of genotypic frequencies and genotypes suggests that two types of isolating mechanisms are operative in this *Geomys* contact zone. The first is a pre-mating isolating mechanism that results in matings between female *major* and male

knoxjonesi being reduced below any detectable measures with our methods (Baker et al., 1989; Bradley et al., 1991a). The second has been interpreted as being a post-mating isolating mechanism that results in F₁ and F₁-like individuals having low reproductive success. It is probable that male F₁ individuals are sterile (Haldane, 1922; Pembleton and Baker, 1978; Baker et al., 1989; Bradley et al., 1991a). Previous workers on this contact zone have interpreted their results as indicating that this narrow hybrid zone is maintained by the dynamic-equilibrium model as outlined by Bigelow (1965) and Moore (1977).

The results of the contiguous clustering analysis differs from those of the *Peromyscus leucopus* hybrid zone by having more discrete clusters and a higher degree of character correlation. It would be convenient to attribute these differences to the biological features that maintain these zones. Table 3 lists some of the characteristics of the *Geomys* and *Peromyscus* hybrid zones. However, these differences may be an artifact of the dif-

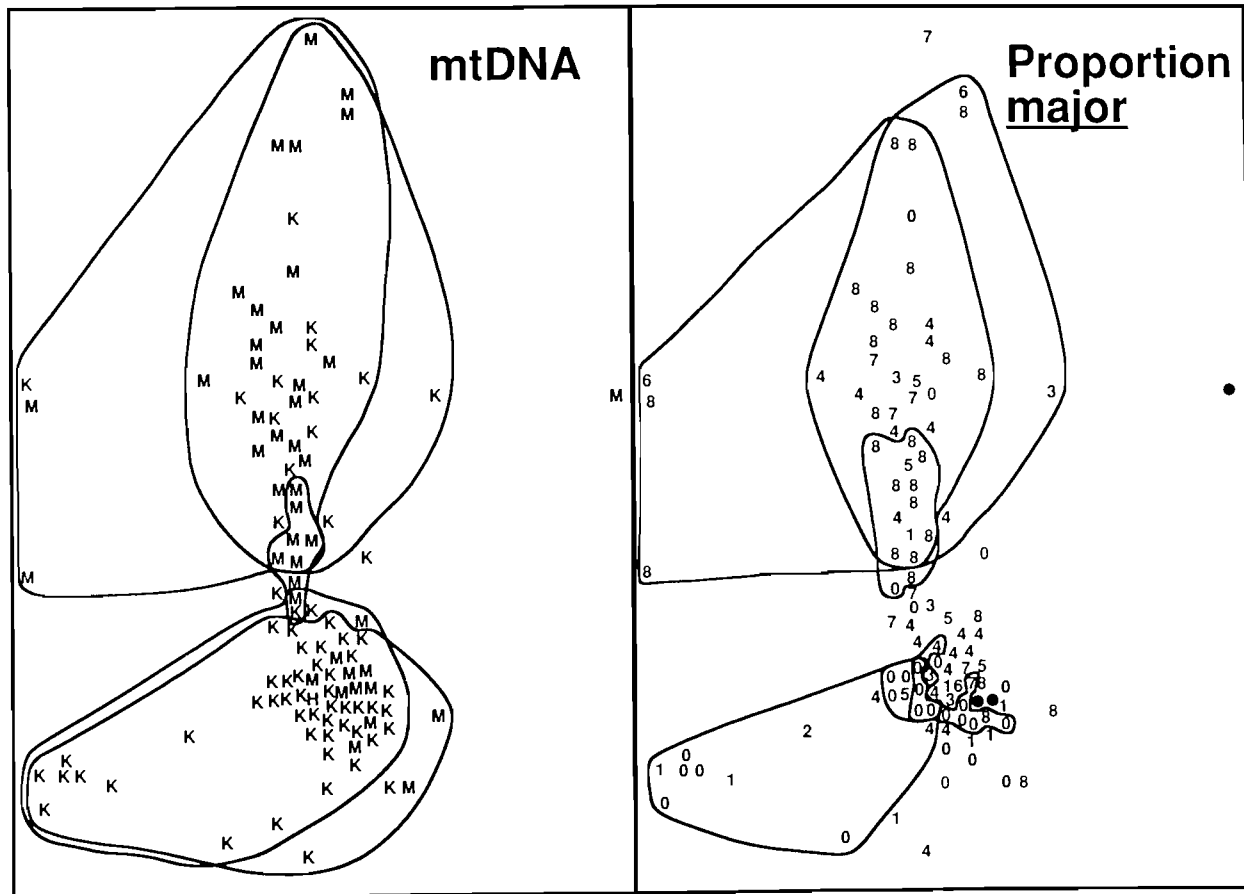


Fig. 5. Graphical representation of the position of individuals from a hybrid zone between *Geomys bursarius major* and *G. knoxjonesi*. Contiguous clustering analyses were performed on the mitochondrial DNA haplotype and the proportion of an individual's genome that was characteristic of the *G. b. major* genome. Solid lines represent contiguous clusters of individuals from the cross entropy X^2 ($P < 0.05$). Some distortion of the ellipses has occurred to make the figure legible.

ference in scale of sampling on which these two studies were based. The *P. leucopus* sample consisted of a 0.571 km \times 0.271-km grid in which about 95% of all individuals were hybrids with no northeastern types and only three southwestern types present in the sample. Therefore, clusters of pure parental types were not possible. Alternatively the *Geomys* sample was collected on a grid of 3 km by 6 km and contained 26.4% parental *major*, 22.7% parental *knoxjonesi*, and 50.9% hybrids. The *Geomys* grid clearly extends beyond the boundaries of a 20/80 definition of a hybrid zone. To have a comparable contiguous clustering analysis of the *P. leucopus* hybrid zone the grid would need to be approximately 60 km by 30 km in size. Alternatively we could restrict our analysis to a grid size reflective of the percentage of the hybrid zone examined for *P. leucopus*. However, this would result in so few individual *Geomys* sampled that the analysis would be insignificant.

Even if we examined a larger area for the *P. leucopus* grid we would anticipate that our results would

still support the hybrid equilibrium model as the best descriptor of the maintenance of this hybrid zone. The dynamic equilibrium model implies the evolution of isolating mechanisms or other genetic features to reduce the fitness of hybrids. The high percentage of backcross individuals in our sample of *P. leucopus* clearly refutes such processes. Also the hybrid superiority model involves the interaction between the environment and individuals with hybrid genotypes over a narrow ecological transition, again not evident from our samples of *P. leucopus*. In fact, both the dynamic equilibrium and the hybrid superiority models under most circumstances would produce relatively narrow hybrid zones and therefore intensive sampling in the center of such narrow zones may be impractical for higher vertebrates. Alternatively, if the pocket gopher sample was reduced to fit the 80/20 criterion, the concordance of clusters would have remained as predicted by Van Den Bussche et al. (1993) for the dynamic-equilibrium model.

Table 3. Relative comparison of the *Geomys* hybrid zone and the previously described *Peromyscus leucopus* hybrid zone (Van Den Bussche et al., 1993).

Characteristic	<i>Geomys</i>	<i>Peromyscus</i>
Habits	fossorial	scansorial
Social Status	solitary	communal
Territoriality	very high	high
Fecundity	low	moderate
80/20 definition	3 km	30.6 km
Pre-mating isolating mechanism	yes	no
Post-mating isolating mechanism	yes	yes
Fixed alleles	distinguish pure parentals	do not distinguish pure parental
Chromosomal races	yes	yes
Type of chromosomal variation	different diploid numbers, and chromosomal rearrangements (mostly heterochromatic)	Races distinguished by three presumed pericentric inversions (fixed)
Ecological change	abrupt - < 3 km	extends over 50 km
Genetic markers	RFLPs, heterochromatic markers distinguish zone	RFLPs, heterochromatic markers distinguish zone
Infertility	strong evidence in hybrids	little evidence in hybrids
Zone defined best by	dynamic equilibrium model	hybrid sink model

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