PRELIMINARY CHROMOSOME STUDIES OF SOME NEVADA TEST SITE LIZARDS

William P. Hall III

INTRODUCTION

This report will discuss results of a study of chromosome number and morphology of some Nevada Test Site lizards. The work reported is a portion of the UCLA Rock Valley Project, a long term study of the effects of chronic, low-level, gamma radiation on a natural desert community. The Rock Valley Project is supported by the USAEC Division of Biology and Medicine. The chromosome study was initiated to provide background data for karyotypic analysis of natural lizard populations being subjected to continuing radiation exposure.

The data presented in this discussion resulted from preliminary experiments designed to assess the usefulness of testes squash preparations for precise analysis of lizard karyotypes.

My chromosome studies were initially suggested by Don Hunsaker and have been given subsequent encouragement by Richard Etheridge and other faculty members of San Diego State College.

MATERIALS AND METHODS

Iguanid lizards from the Nevada Test Site, located in Nye County, Nevada were used for this study. Standard aceto-orcein stained squash preparations were made from testes following hypotonic pretreatment. The slides were made permanent by the dry ice quick freeze methods, and were examined with phase-contrast optics. Most determinations were made from meiotic figures, and were confirmed by mitotic counts when possible. Many figures were counted from several individuals of each species.

RESULTS

The squash preparations were inadequate for detailed analysis of lizard karyotypes. However, the testes squash technique proved to be simple and useful for survey work. I believe the chromosome numbers and rough karyotypes determined from these preparations are reasonably accurate. I don't think they are definitive, though, because of the many minute elements in the figures; which are further confused by possible heteromorphic sex chromosomes. The first three slides will illustrate. Slide number one shows three reasonably good meiotic figures from *Sceloporus jarrovi*, a species with probable heteromorphic chromosomes. The next slide shows a mitotic figure from *Crotaphytus wislizeni*, where there is possibly an unpaired chromosome. I show slide number three for comparative purposes. Notice chromosome pair number seven, probably the sex chromosomes. This is a rough photographic karyotype, constructed from one of my first successful trials of the peripheral blood culture technique. This will be used for future karyotypic studies.

Table Number One (next slide) presents the cytogenetic results obtained from my studies of Test Site materials. All species have six pairs of metacentric macrochromosomes, and a variable number of structurally undefined microchromosomes. When combined with chromosome data for other iguanid species, the karyotypic variation suggests some theoretically interesting implications.

DISCUSSION

Table number two lists all the iguanid chromosome data I have been able to collect. The macrochromosome compliment remains constant in all species. Sceloporines, as recently defined by Richard Etheridge, have a reduced number of microchromosomes, varying between five and eleven pairs. All other iguanids have 12 pairs. Sceloporines are also notable, among the iguanids, for their large number of species and their adaptive radiation entering all North American terrestrial, and many arboreal, lizard habitats. The correlation between this, and the karyotypic variation, suggests they may be causally related.

However, before I discuss this possible relationship, I would like to comment on the probable mechanism of the sceloporines karyotypic variation, and about my concepts of the species.

Robert Matthey, and M. J. D. White have called the reciprocal, wholearm translocation of long and short arms, from two non-homologous, achrocentric chromosomes, a centric fusion. The chromosomes resulting from this fusion consist of metacentric, and a centromere practically bare of genetic material. In heterozygote meiosis, the long achrocentric arms would pair with the metacentric's arms, leaving the bare centromere unpaired. It would soon be lost from the population under these conditions, and its absence would have little, if any effect on the population's survival.

The two achrocentrics, and the fused metacentric would also be expected to encounter division problems in meiosis. Two of the chromosomes would certainly pass to opposite poles. However, the third would be expected to pass to one pole more-or-less at random. Therefore, the heterozygote would produce a significant number of non-disjunction gametes, causing an appreciable sterility. Since the fusion initially would be present in just one individual, partial heterozygote sterility would preclude fixation of a centric fusion in a large population. However, fusions would have a definite probability of fixation in very small, isolated populations. This probability would depend on the number of interbreeding individuals, and on the percentage of heterozygote sterility. The reverse process to centric fusion is quite improbable, since it is believed to require the production of a new centromere.

On this basis, the non-sceloporine iguanid karyotype is probably primative. The six pairs of metacentric macrochromosomes, and the twelve pairs of presumably achrocentric microchromosomes, would have 48 chromosome arms. Matthey believes this is the fundamental number for alarge group of lizards, including the family Iguanidae. The sceloporine karyotypes can be derived from this primative form, if some of their microchromosomes are assumed to be metacentric. Except for the supposed sex-chromosomes, the detailed karyotype of Sceloporus cyanogenys, presented earlier, meets this criterion.

My species concepts rely heavily on the works of Andrewartha and Birch, Mayr, and Ford. A species may be thought of as a genepool -- defined as the total number of genetic loci and alleles, carried by potentially interbreeding organisms. This pool exists in a multi-dimensional environment of independent variables, each variable having selective effects on the distribution of the pool's genes. The genes also have selective effects on each other, since all must act in unison for an organism's growth and function.

A species' distribution depends on the adaptive limits of its genepool, and is determined by the peaks and valleys in the environmental coordinates. In nature this produces a number of local populations, existing in semi-isolation, which are the units of the genepool.

The number of individuals in local populations depends on selective values of environmental coordinates, and on adaptability of the local segment of the species' genepool. This adaptability is a function of the distribution of genes present, and of new mutations arising in the pool.

Several important factors influence genes adapting a local population to a peripheral environment. Mutations, adaptive to stresses of the peripheral environment will lack a history of adaptation to the existing genes of the pool. Because of this they would probably be at a selective disadvantage to older alleles because the mutations would operate less efficiently with the genepool. However, environmental selection by continued high stress would probably be sufficient to counterbalance negative genetic selection against an adaptive mutation, even though it lowered the organism's efficiency. The continued stress conditions would also favor co-adaptive mutations at other loci, to regain lost efficiency. A history of high environmental stress, insufficient for extinction, and continued environmental isolation might eventually lead to the production of a new species. However, enough gene changes must be accumulated to produce genetic or ecological isolation when the populations meet, or the differences will break down. This is a slow and improbable process, depending on the accumulation of many mutations in a complete isolated genepool, under continued less-than-lethal environmental stresses.

One more, very important factor influences peripheral adaptability. This is temporal variation in environmental coordinates. Local populations expand when environmental stresses relax. Under these conditions, there will be little, or possibly even negative, environmental selection for peripheral mutations. And the genetic selection against them will continue unabated. In addition to the numerical expansion of local populations under optimum conditions, individual migrations will be extensive. These migrants may inject whole gene complexes, with a long history of co-adaptation, into peripheral populations, to further disadvantage peripheral mutations under relaxed stress. At the other extreme, periods of maximum environmental stress, would extinguish a species' most peripheral, and probably, most isolated populations. These temporal variations, combined with the genetic mechanisms mentioned, are probably responsible for maintaining the integrity and continuity of a species within a well defined range against the disruptive effects of selection.

CONCLUSIONS

To recapitulate the most important ideas already stated: Fixation of a centric fusion is possible only in very small, isolated populations. Only the most peripheral surviving populations of a species are likely to meet this criterion. Also, these should be among the most genetically divergent populations. Individuals heterozygous for a centric fusion are probably semi-sterile. Once a fusion becomes fixed in a population, heterozygotes are likely only when environmental factors are optimum and migrating individuals are encountered.

The fixation of a centric fusion in a peripheral population might lead to the establishment of a new species as follows: Continued or repeated stress periods would be expected to shift the adaptive parameters of the peripheral genepool away from parameters of central gene pools. During periods of optimum environment, peripheral and central populations would expand and make contact, at least through individual migrations. However, within the peripheral population, members would outnumber migrating individuals, and would form semi-sterile hybrids with them, if they mated. Because of the semi-sterility of interpopulation hybrids, many central gene complexes would be barred from the peripheral pool. Also, genes leading to interpopulation hibridization would be selectively lost. Genes would be selectively favored that provided ecological or behavioral isolation. In this case, environmental fluctuations would amplify divergence and encourage the invasion of new environmental niches by the peripheral gene pool, instead of operating to swamp its divergent characters as would take place otherwise. This chromosomal mechanism could conceivably produce a new species within a period of a few environmental cycles, because of continued high selection for divergent characters and isolating mechanisms under all environmental conditions. Non-chromosomal mechanisms of speciation would most likely take thousands of generations.

This chromosomal mechanism could explain the sceloporine's diversity, in contrast to the conservative or specialized nature of most other iguanids. As far as I know, within the sceloporines, small genera with fairly restricted habitats, show little chromosomal variability. Sceloporus, with a great karyotypic variability, has invaded most North American lizard habitats with one or more species.

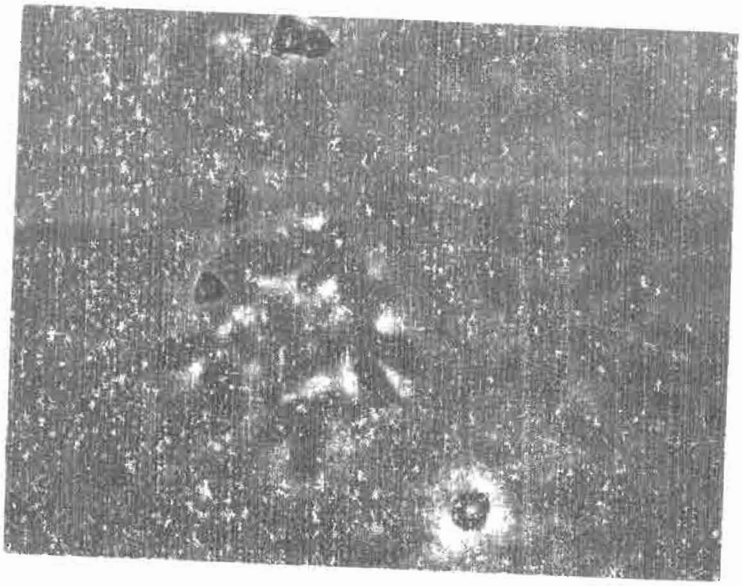
I think the existance of a mechanism of speciation involving centric fusion is reasonable and likely, since it would have selective value. In peripheral populations the incidence of a low percentage of hybrid non-disjunction would be adaptive. This would reduce the number of environmentally maladaptive genes entering peripheral genepools during optimum conditions, allowing better survival during environmental extremes. The effect would be enhanced by selection. When it was sufficient for production of a new species, this new species would have mechanisms to spawn further species more readily. Groups lacking the mechanisms would be slow to occupy new environments, and hence slow to adapt to major environmental changes. Groups using this speciation mechanism would frequently be trying new environments, while less active groups went to extinction because their optimum environment vanishes, and other possible niches were already filled by more active groups.. The adaptive radiation would eventually be limited by the number of possible centric fusions, but in the meantime, many new species would be produced.

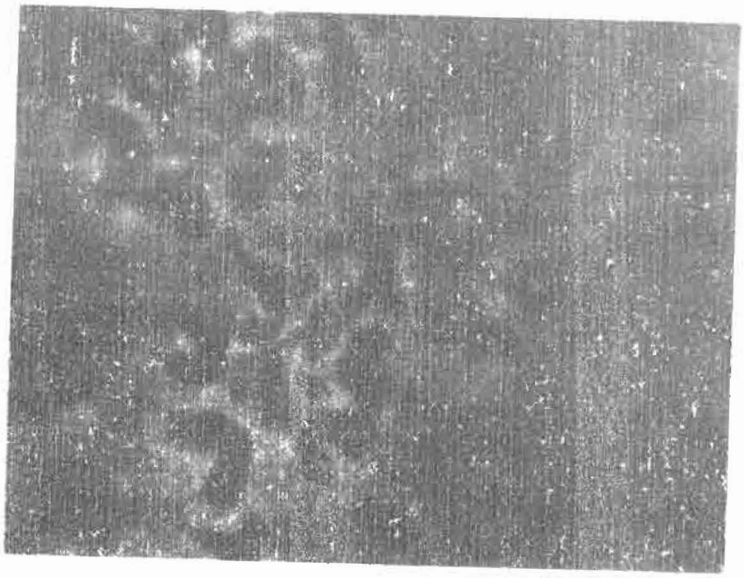
The iguanids were apparently limited by a discontinuity in chromosome size between the 24 longest arms and 24 shortest arms, ancestral forms having already exploited this variation. Possibly some mutation in primative sceloporine lizards made centric fusion of the michrochromosomes easier, and opened the way for a new adaptive radiation based upon it.

Several aspects of this hypothesis are subject to experimental verification, or at least support. As time and money is available, I am making a detailed karyotypic analysis of the undulatus group of Sceloporus in an attempt to gain a better understanding of the distribution and mechanisms of the karyotypic variation observed in this group. This study should either support or deny basic assumptions of the hypothesis.

SUMMARY

To summarize: In this report I discussed my assessment of a testes squash technique for detailed analysis of lizard karyotypes. Although the technique was unsuitable for the stated purpose, combined with other data, it provided enough chromosome information to suggest some possibly important ideas concerning the origin and evolution of species. A chromosomal isolating mechanism may have been important in the adaptive radiation of new species in the lizard family Iguanidae. The fixation of centric fusions in peripheral populations would encourage speciation by protecting peripherally adapted gene pools from being swamped by centarally adapted gene complexes, and by further encouraging the selection of other isolating characters, during optimum conditions.





S cyanogenys #1 No Locality Data 104

Slide No 215 55.7 x 20.5 x 2666

Peripheral Blood Culture

80% Medium 199 20% Chicken Serum