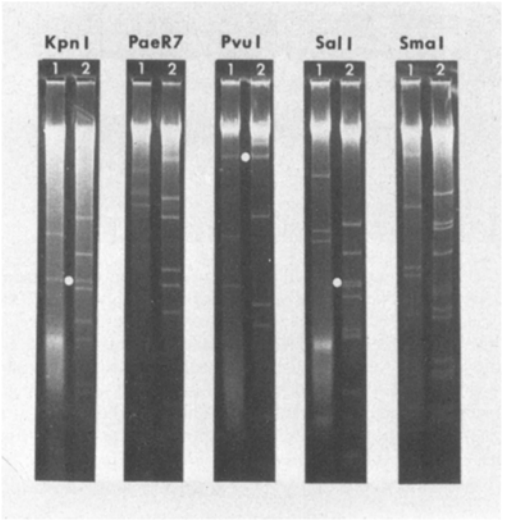

Electrophoretic patterns of (1) MAd-FL and (2) MAd-K87 DNAs after cleavage with five restriction endonucleases. Electrophoresis of DNA fragments was carried out in a 1 % agarose slab gel at 50 V for 16 h. DNA in the gel was stained with ethidium bromide and photographed under ultraviolet light illumination. Comigrating MAd DNA fragments are indicated by white dots.

Only one species of murine adenovirus (MAd) is until now recognized in the literature[2, 3]. MAd prototype strains FL and K87 differ, however, in pathogenicity and tissue tropism[4–7]. Serological tests also indicate that the two viruses are antigenically distinct[8–10]. Under these circumstances, it seemed interesting to use restriction endonuclease analysis of viral DNA as a means to further differentiate the MAd strains.

The FL and K87 viruses were propagated in primary mouse kidney cells and embryonic mouse cells, respectively, as described earlier[9]. Viral DNA was extracted from MAd-infected cells by the SDS-pronase-phenol method developed earlier for canine adenoviruses[11]. Aliquots containing about 0.5 µg of MAd-FL or MAd-K87 DNA were digested for 2 h at 37 °C under appropriate buffer conditions with restriction enzymes (5–10 units) either prepared in our laboratory (*Pae*R7) or purchased from Bethesda Research Laboratories (*Kpn* I, *Pvu* I, *Sal* I and *Sma* I). Digestion products were resolved by electrophoresis for 16 h at 50 V in a 1 % agarose gel as described previously[12]. DNA in the gel was stained with ethidium bromide (1 µg/ml) and photographed under UV light illumination.

According to the restriction profiles shown in the figure, the FL and K87 strains represent two distinct species of MAd. The number of *Kpn* I, *Pae* R7, *Pvu* I, *Sal* I and *Sma* I restriction fragments is, effectively, different for the two viruses. MAd-FL and MAd-K87 DNA fragments rarely comigrate in the gel (white dots). Hexanucleotide sequences recognized by the five restriction endonucleases used in this work are thus located at very distinct positions on each type of MAd DNA molecules.

Together with biological and immunological differences already found[4–10], our results confirm the existence of at least two distinct adenovirus species in the mouse. The size of each restriction fragment in the gel has now been evaluated using the 1 kilobase ladder from Bethesda Research Laboratories as a molecular weight marker. Results indicate that the MAd-FL and MAd-K87 genomes comprise 32 and 31 kilobase pairs, respectively. Similar values were published earlier by Larsen et al.[13, 14] for MAd-FL DNA.

The usefulness of restriction endonuclease analysis in the identification and classification of DNA viruses is also confirmed. This sensitive and specific technique was recently used in our laboratory to distinguish between canine adenovirus types 1 and 2[15, 16]. With the present results, we can only support the concept of genome typing for species distinction. As suggested earlier by Whetstone[17], the definition of an adenovirus species should include both serotype and genotype. In this perspective, the discovery of new adenovirus species in rodents is more than probable.

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Karyotypic differentiation in an agamid lizard, *Japalura swinhonis swinhonis*[1]

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Summary. Three populations of a Taiwanese agamid lizard *Japalura swinhonis swinhonis* exhibit karyotypes distinct from each other. Lizards from the northern area have $2n = 46$ all acrocentric chromosomes, whereas animals from two localities in the central region possess $2n = 36$ and $2n = 40$ chromosomes, respectively, including several biarmed elements.
Key words. Japalura s. swinhonis; Reptilia; Agamidae; karyotype.

The Taiwanese members of the genus *Japalura* were taxonomically reviewed by Liang and Wang[2] on the basis of morphological characters. They combined all the populations within this island into a single species comprising three subspecies: *J. swinhonis swinhonis, J. s. mitsukurii,* and *J. s. formosensis*. Of these, *J. s. swinhonis* is known from both the northern lowland, and the central mountainous region[2,3]. In the present paper, I report on the considerable karyotypic differentiation observed among three populations assigned to *J. s. swinhonis*.

Materials and methods. Fourteen lizards, identified as *J. s. swinhonis* after Liang and Wang[2], were karyotyped by a bone-marrow air-dry method following Ota et al.[4]. Localities and the numbers of lizards examined were: Taipei (fig. 1 A), 4 males and 1 female; Lushan (fig. 1 B), five males; Chitou (fig. 1 C), four males. The karyotype of each population was determined from over 35 well-spread cells, and described following the terminology of Green et al.[5].

Results. No heteromorphic pairs were evident in the karyotypes observed. Lizards from Taipei possessed a karyotype comprising $2n = 46$ all acrocentric chromosomes in a graded series (fig. 2 A), and the N.F. value was therefore calculated as 46. Karyotypes of the specimens from Lushan and Chitou differed from that of the former in including several biarmed elements, as well as in exhibiting divisions into size-groups. Lizards from Lusan possessed a karyotype consisting of $2n = 36$ chromosomes forming two distinct size-groups. The larger group accommodated five pairs, of which pairs 1, 3, 4 and 5 were metacentric, and pair 2 submetacentric. Pairs 6 to 18, all belonging to the smaller group, appeared to be acrocentric elements (fig. 2 B). Thus, the N.F. value was calculated as 46. Specimens from Chitou, however, had a karyotype comprising $2n = 40$ chromosomes forming three discontinuous size groups. The largest group consisted of two pairs, one metacentric and one submetacentric. The intermediate group comprised seven pairs, of which pair 4 was acrocentric, pair 6 submetacentric, and the remainder subtelocentric elements. Of the chromosomes belonging to the smallest group, pairs 10–16 appeared as biarmed and the others as uniarmed elements (fig. 2 C). Therefore, the N.F. value was calculated as 70.

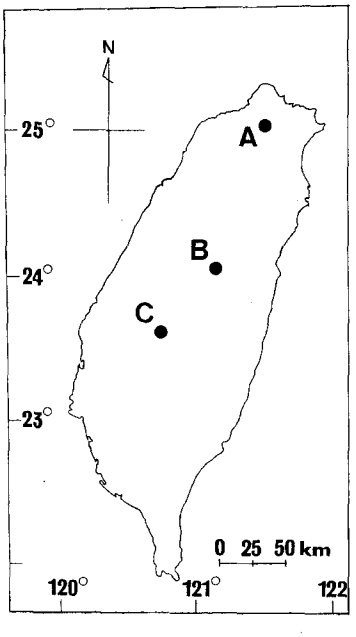

Figure 1. Sampling localities of the present materials. *A* Taipei; *B* Lushan; *C* Chitou.

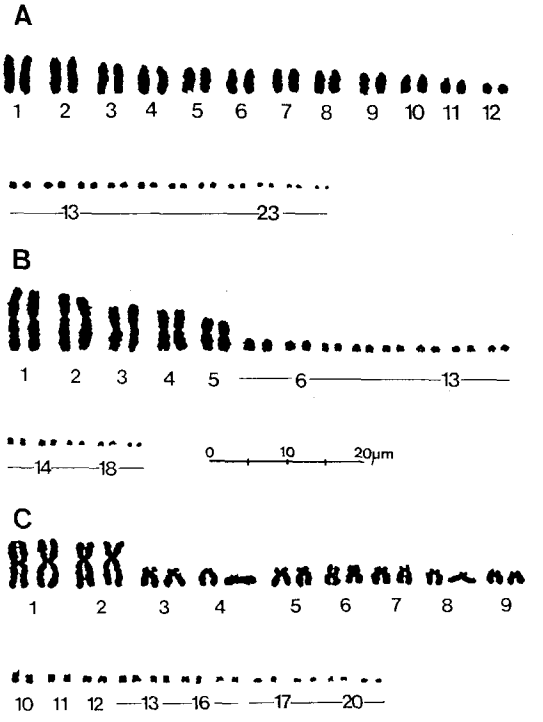

Figure 2. Karyotypes of *Japalura s. swinhonis* from *A* Taipei, *B* Lushan, and *C* Chitou.

Discussion. Recently, chromosomal variation within a morphologically defined species or species complex has been reported for several lacertilian families (Gekkonidae, Xantusiidae, Iguanidae, Anniellidae, and Scincidae)[6]. The present study revealed that such variation also exists in the family Agamidae.

The karyotype of this family has been little studied, considering the morphological and ecological diversities of the group[7,8]. Of 20 species assigned to the genus *Japalura*[2,9,10], only three have hitherto been karyotyped: *J. swinhonis*[11], *J. polygonata*[12], and *J. varcoae*[13].

The karyotype of the sample from Taipei is identical with those previously reported for *Japalura swinhonis* from northern Taiwan and *J. polygonata* from Okinawa Island[11,12]. However, the karyotype of the sample from Lushan most resembles that of *J. varcoae* from Yunnan, which consists of 12 large biarmed chromosomes and 22 micro-chromosomes[13]. All these karyotypes share an N.F. value of 46. On the other hand, the N.F. value (70) in the karyotype of the sample from Chitou is markedly larger than that of any other diploid species of the family Agamidae previously karyotyped[6,14].

Li et al.[13] assumed the karyotype of *Japalura* to be conservative in terms of arm number, and postulated that the known karyotypes of *J. swinhonis* and *J. polygonata* had derived from a karyotype including several biarmed elements through a series of centric fissions. However, the karyotype of the sample from Chitou represents a case inconsistent with the above assumption, and implies the occurrence of chromosomal reorganization by other modes involving changes in arm numbers (such as pericentric inversion, addition, or deletion of chromatin), although further detailed analyses with chromosome-banding techniques are required to outline the actual process of karyological divergence in *Japalura*.

The present results seem to indicate reproductive isolation among the three populations presently assigned to *J. s. swin-*

honis[2, 3]; it is highly probable that each of these populations represents a good biological species. Thus, further detailed morphological comparisons among these populations are also required in order to revise the taxonomy of the Taiwanese *Japalura*.

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The karyotype and genome structure of the pirate perch *Aphredoderus sayanus* (Aphredoderidae: Teleostei)

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Summary. The standard karyotype, genome size (DNA content), and genomic DNA base composition and distribution of the relict paracanthopterygian fish, *Aphredoderus sayanus*, were investigated. Several features of the *A. sayanus* genome appear to be derived rather than primitive conditions. These include a large number (at least 10 pairs) of bi-armed chromosomes, a low genome size, and high DNA asymmetry. This may indicate that *A. sayanus* is not a typical paracanthopterygian fish in terms of its genome structure.
Key words. Karyotype; genome size; DNA base composition; relict fish.

The pirate perch, *Aphredoderus sayanus*, is the only living member of the family Aphredoderidae, and is one of the few freshwater forms in the teleost superorder Paracanthopterygii[1]. The species is endemic to North America and is found primarily in the lowlands of the Atlantic and Gulf slopes and in the Mississippi Valley[2]. Relatively little is known about its biology, although there are published data on pirate perch habitats, food, growth, and reproduction[2, 3]. Evolutionarily, *A. sayanus* is a living relict and presumably represents one of the remnants of an ancient ichthyofauna that occupied the Mississippi Valley prior to the ancestors of most modern day North American fishes[4]. Fossil genera related to *Aphredoderus* are known in North America from as early as the Oligocene[5].

In this note, the standard karyotype, genome size (DNA content), and genomic DNA base composition and distribution of *A. sayanus* are reported. The purposes of the study were to obtain basic genetic information on a poorly known species, and to examine the chromosomal and genomic structure of a paracanthopterygian fish. The latter are not well known genetically since most paracanthopterygian species are marine and difficult to obtain alive. Paracanthopterygians are of systematic interest since they are the putative sister group to the Acanthopterygii, the largest and most diverse of all presumably monophyletic teleost groups[6, 7].

The *A. sayanus* specimens examined in the study were collected by seine from an unnamed tributary of the Navasota River near College Station, Texas. The specimens were returned live to our laboratory and maintained in aerated aquaria until sacrificed. The methods used to prepare, stain, and photograph metaphase chromosomes followed Gold[8]. Genome sizes of two individuals were determined by flow cytometry of erythrocyte nuclei and sperm following the methods of Bickham et al.[9] and using chicken erythrocyte nuclei as the internal standard. Genomic DNA base composition and differential melting rate profiles were generated via thermal denaturation of visceral tissue DNA isolated from two individuals following the methods of Mandel and Marmur[10].

The standard karyotype of *A. sayanus* (fig. 1) contains $2n = 48$ chromosomes as determined from over 150 metaphases taken from four specimens. Fundamental arm number (NF) estimates ranged from 68 to 72 as measured from six different karyotypes. One pair, the largest in the complement (cf fig. 1), was clearly heteromorphic in two of the specimens and may indicate the existence of morphologically differentiated sex chromosomes. These two specimens were male; the other two specimens karyotyped were juveniles and could not be identified as to sex. This means that the heteromorphism detected could well be autosomal. The occurrence of bi-armed chromosomes in the *A. sayanus* karyotype is probably a derived rather than primitive condition. Over 1800 teleost species have been karyotyped, including 15 species from the superorder Paracanthopterygii and over 900 species from the superorder Acanthopterygii[11]. A diploid karyotype of 48 acrocentric chromosomes (i.e., $2n = 48$, $NF = 48$) has been found in most of the major teleost groups, and is the predominant karyotype in the most advanced group, the Acanthopterygii. On this basis, Ohno[12] and others have suggested that the $2n = 48$, $NF = 48$ condition is primitive for teleosts. *A. sayanus* possesses a diploid number of 48 chromosomes, but at least 10 pairs of chromosomes are bi-armed (meta- or submetacentric) and one of these may possibly represent a sex chromosome heteromor-