lor's deductions (*2*). In this connection, it is also interesting that condensed and transcriptionally inactive heterochromatin constitutes about 90 percent of the chromatin in a typical mammalian cell (*19, 20*) and that virtually all break points induced by x-irradiation in mitotic chromosomes appear in their G–light-banded regions (*21*).

Normal cells irradiated with x-rays undergo a delay in the initiation of DNA synthesis, but A-T cells do not (*22–24*). It has been suggested that the delay occurs because initiation of DNA synthesis cannot take place while damaged DNA is undergoing repair and that the chromatin structure of A-T cells presents itself as a suboptimal substrate for such repair (*5, 23*). Our experiments with noncycling cells show that progression into the *S* phase is not a prerequisite for the increased frequency of chromosome fragments that appear in mitosis after ataxia cells are irradiated in the G_1 or G_0 phase (*2, 24*).

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12. Curves of the form $Y_T = Ae^{-ct} + Be^{-dt}$ were also fitted to the gross data (total PCC's plus fragments) for each experiment. In no case was the value of d significantly different from zero. For this reason, and because Y_T (the total number of PCC's plus fragments) would not be expected to decrease below that for unirradiated cells, even after very long incubation times, the exponent of the second term was set at zero and the net frequencies of fragments, Y (weighted according to the gross data), were fitted to the simplified expression.
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Diploid-Triploid Mosaicism: An Unusual Phenomenon in Side-Necked Turtles (*Platemys platycephala*)

Abstract. *Diploid and diploid-triploid mosaic individuals of* Platemys platycephala *were found in natural populations. In mosaic specimens, the blood, spleen, liver, and testis contained both diploid and triploid cells. The ratio of triploid to diploid cells was more variable among individuals than among somatic tissues within an individual. Only diploid cells underwent meiosis in males; haploid gametes were produced. There appears to be geographic variation for mosaicism in that only diploids were found in Bolivia, whereas diploids and diploid-triploid mosaics occurred in Surinam.*

Triploidy is rare in vertebrates but occurs as a natural condition in fishes (*1*), amphibians (*2*), and reptiles. Populations of triploid individuals are known in the lizard families Teiidae, Gekkonidae, and Agamidae (*3*), but have not been reported for any of the other major reptile groups (snakes, crocodilians, turtles, or the tuatara). Triploid vertebrate populations are invariably unisexual (all fe-

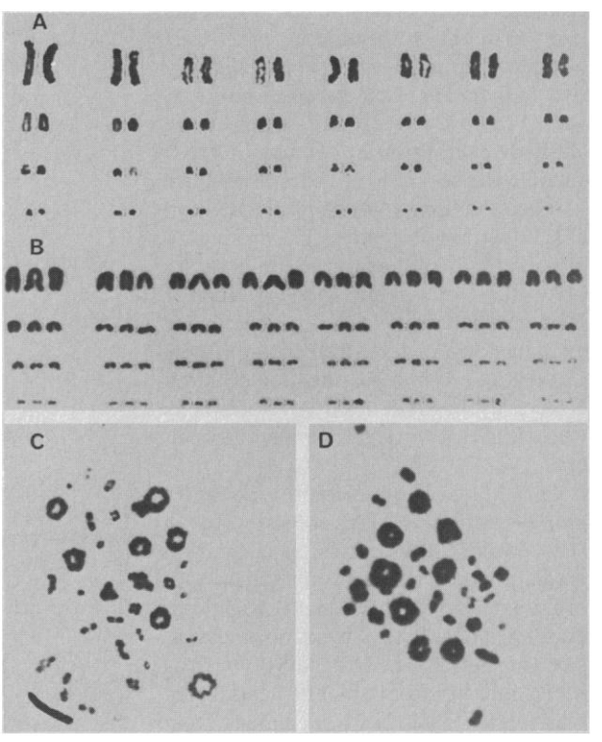

Fig. 1. Mitotic and meiotic chromosomes from *P. platycephala*. (A) Diploid ($2n = 64$) mitotic karyotype from the testis of a specimen from Bolivia. (B) Triploid ($3n = 96$) mitotic karyotype from the spleen of a diploid-triploid mosaic individual (AK1227). (C) Diakinesis in a diploid male from Bolivia with 32 bivalents. (D) Diakinesis in a diploid-triploid mosaic male (AK1227) with 32 bivalents.

Table 1. Percentage of triploid cells in tissues from individual diploid-triploid mosaic *P. platycephala*. Flow cytometric data represent the total number of cells in the 2C and 3C peaks except where otherwise noted.

Individual	Tissue	Number of cells		Triploid cells (%)	Technique*
		Diploid	Triploid		
AK1227	Spleen	1	20	95	K
	Testis†	8	2	20	K
AK6630	Testis	525,460	157,779	23	FCM
AK6609	Blood	886	1395‡	61	FCM
	Spleen	1	9	90	K
AK6626	Blood	322,958	71,950	18	FCM
	Liver	711,741	187,033	21	FCM
	Testis	73,200	32,380	31	FCM

*K, karyotype; FCM, flow cytometry. †Only mitotic metaphases were scored. ‡Values represent mean channel scores only rather than total cells in the entire peak.

male) and reproduce by way of parthenogenesis, gynogenesis, or hybridogenesis (*4*).

The spontaneous production of triploids occurs in a variety of vertebrates. These are usually considered to be the result of an unreduced gamete fusing with a normal haploid one (*5*). These triploids are viable in some vertebrates (*6*) but are not viable in man (*7*). A triploid male lizard was viable but produced unreduced gametes (*8*). Diploid-triploid mosaicism or chimerism occurs spontaneously in mammals and has been reported for a single case in a lizard. Such individuals may survive to adult age (*9*). Mosaicism is a rare event and not thought to be involved in the process of speciation or adaptive evolution.

We have found diploid-triploid mosaicism maintained in a naturally occurring population of South American turtles, *Platemys platycephala* (family Chelidae). In most cases, karyotypes were obtained directly from spleen and testis (*10*). In addition, leukocyte and heart fibroblast cultures were used to study two individuals. Cell suspensions from whole blood, liver, and testis were washed several times in Hanks buffered saline solution (BSS), fixed in a mixture of equal volumes of Hanks BBS and absolute ethanol, stained in chromomycin A_3 (*11*), and analyzed on a Leitz MPV flow cytometer with a Nuclear Data pulse height analyzer. Specimens examined were deposited in the Texas Cooperative Wildlife Collection or at the University of Utah (*12*). All specimens were obtained through commercial dealers.

The diploid mitotic karyotype of *P. platycephala* (Fig. 1A) consisted of 64 acrocentric chromosomes ranging from medium-sized macrochromosomes (approximately 5 μm in length) to microchromosomes. There were no apparent sex chromosomes. This karyotype was found in cells from spleen, heart fibroblasts, and testis in 21 specimens from Bolivia and is identical to that found in a Brazilian specimen (*13*). Meiocytes from reproductively active males had 32 bivalents at diakinesis (Fig. 1C) and 32 chromosomes at prophase of the second meiotic division. No evidence of triploidy was found in specimens from Bolivia.

One male individual of unknown geographic origin (AK1227) had a karyotype composed of 96 acrocentric chromosomes ranging from medium-sized macrochromosomes to microchromosomes (Fig. 1B). Cells with this karyotype were found in the spleen and testis. This karyotype is identical to that reported from lymphocyte cultures of two individuals of *P. platycephala* (*14*). The majority of mitotic metaphases in the testis of AK1227 had 64 chromosomes, whereas nearly all of the spleen cells were triploid (Table 1). Meiocytes had 32 bivalents at diakinesis (Fig. 1D) and 32 chromosomes in prophase cells at the second meiotic division.

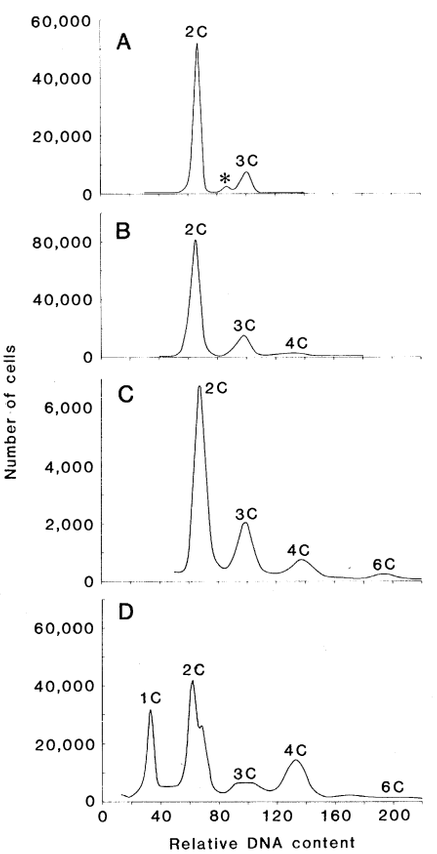

Fig. 2. DNA histograms from two individual diploid-triploid mosaic *P. platycephala*. Blood from AK6626 (A) shows two major peaks corresponding to 2C (diploid) and 3C (triploid) cells. A smaller peak between the major peaks (asterisk) might represent a population of modified triploid cells but this peak was not seen in liver and testis. (B) Liver from AK6626. (C) Testis of AK6626. (D) Testis from AK6630.

Three male specimens from Surinam were diploid-triploid mosaics. One individual (AK6609) was predominantly triploid in both spleen and blood (Table 1). Another (AK6626) was predominantly diploid in blood, liver, and testis (Fig. 2, A and B, and Table 1). The distribution of cells from the liver of AK6626 (Fig. 2B) showed major peaks at the 2C and 3C positions and minor peaks at the 4C and 6C values (*15*). The histogram of cells from the testis of AK6626 (Fig. 2C) had two major peaks (2C and 3C) and two minor peaks (4C and 6C). This individual was not in reproductive condition. All three tissues of AK6626 were predominantly diploid but the proportion of diploid to triploid cells was variable (Table 1). The third individual (AK6630) was predominantly diploid in the testis (Fig. 2D and Table 1). Cells from the testis of AK6630 were distributed as four major peaks corresponding to 1C, 2C, 3C, and 4C values and a small 6C peak was also present. This individual was in reproductive condition and produced haploid (1C) gametes. The 2C peak (diploid) was actually double with the mean channels of each peak at 62 and 68. The 3C peak (triploid) was low and broad and may correspond to triploid cells derived from both 2C populations. The prominent 4C peak presumably reflects the increased cell division of the diploid populations undergoing meiosis. The triploid cells were not undergoing rapid cell division as indicated by the very small 6C peak.

Flow cytometric analysis of blood from an additional (independently obtained) 16 specimens from Surinam revealed that 11 were diploid, 1 was triploid with no evidence of a 2C peak, and 4 were diploid-triploid mosaics possessing both 2C and 3C peaks (*16*). Mosaic specimens included both males and females. In sum, mosaicism is indicated in 8 out of a total of 19 specimens from Surinam (including the triploid specimen that was mosaic in the liver).

Although turtles are one of the most

intensively studied reptile groups in terms of karyology (*17*), only one report of triploidy has appeared (*14*). Diploid-triploid mosaicism has not been observed in other animals, although cells of certain tissues increase DNA content through endopolyploidy, gene amplification, or polyteny (*18*). Mosaicism in *P. platycephala* appears to be distinct from these phenomona because it is not limited to a specific tissue or cell type.

Our data demonstrate the existence of a diploid-triploid mosaic system in *P. platycephala*. Some individuals were predominantly triploid and others diploid (Table 1). Haploid gametes were produced by both reproductively active mosaic males examined (AK1227 and AK6630), thus indicating the fertility of these individuals. A female specimen (*14*) also was apparently fertile as she laid an egg while in captivity. Thus, triploidy in *P. platycephala* is not associated with parthenogenetic reproduction as is the case in all other normally triploid reptiles, nor is it associated with sterility as occurs in abnormal triploids (*8*).

We believe these mosaic individuals are autopolyploids and not the result of an interspecific hybridization event because (i) the individuals in question are morphologically typical *P. platycephala*, (ii) initial electrophoretic data do not indicate a higher level of heterozygosity in mosaics compared to diploids (*16*), and (iii) the karyotype does not suggest allopolyploidy. *Platemys platycephala* differs from all other members of the family Chelidae in diploid number and chromosome morphology. The other four species of the genus *Platemys* have karyotypes with diploid numbers of approximately 48 to 50 with several pairs of biarmed chromosomes (*19*).

The mechanism by which diploid-triploid mosaicism could be maintained in a sexually reproducing organism is unclear. A zygote must begin as either a diploid or triploid, with subsequent changes in chromosome number occurring in somatic or gonadal tissues during development. In either case, there may be an increase in ploidy to hexaploid and subsequent reduction to diploid or triploid. Furthermore, it is unknown whether the switch from diploid to triploid (or vice versa) occurs only once in development or independently in different tissues.

Mosaics would have the benefits of both sexual reproduction and polyploidy. Autopolyploids possess structural and biochemical properties not found in their diploid progenitors (*5*) and may have larger cell size, and slower development than diploids. They also show an increased ecological adaptation that allows them to exploit habitats that may not be available to diploids. Gene dosage effects might allow genes to be regulated differently in polyploids than in diploids (*5*). The presence of fertile male diploid-triploid mosaic specimens of *P. platycephala* and the presumably fertile female suggests that this cytogenetic system occurs naturally. Geographic variation is indicated as this population (or populations) is in Surinam but only diploids are known from Bolivia and Brazil.

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Isolation of the Gene for a Glycophorin-Binding Protein Implicated in Erythrocyte Invasion by a Malaria Parasite

Abstract. Plasmodium falciparum, *the most lethal of the malarial parasites that infect humans, undergoes three cycles of development in its vertebrate host and elicits stage-specific immune responses. This stage specificity of the immune response has made it difficult to isolate antigens that would be useful in developing a vaccine against malaria. A complementary DNA clone for a glycophorin-binding protein of* Plasmodium falciparum *merozoites has been isolated and characterized. The protein interacts with glycophorin, the erythrocyte receptor, during invasion of the host cell by the parasite. Antigenic determinants of this protein expressed in* Escherichia coli *have been used to produce antibodies to a glycophorin-binding protein. The antibodies show schizont-specific immunofluorescence and react with the merozoite protein. The primary sequence of these determinants reveals a 150-nucleotide tandem-repeating sequence coding for a 50-amino-acid repeat. The characterization of the* Plasmodium falciparum *glycophorin-binding protein represents one approach toward designing serologic agents to block the parasite's development in the vertebrate host.*

The malarial parasite has three cycles of development in its vertebrate host: the hepatic cycle; the asexual blood cycle, which includes the extracellular merozoite and the intraerythrocytic schizont; and the sexual gametocyte cycle. The erythrocyte cycle is responsible for the clinical symptoms of the disease and mortality. One of the obstacles in the development of a vaccine against the malarial parasite is that the immune response in the host is strictly stage specific. Thus, efforts have been directed toward identifying antigens of each stage which, when combined, could be used for vaccination.

The major surface antigen of sporozoites of *Plasmodium falciparum* has been