

## A spontaneous chicken chimaera

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**Summary.** Among hybrids of the inbred lines, a chicken was found to be chimaeric in red blood cells and skin cells and to produce 3 types of sperm cells. This bird could have originated either from the fusion of 2 blastoderms or from the fertilization and fusion of the oocyte and the 2nd polar body.

Experimentally produced avian chimaeras have been described by a number of investigators<sup>2-5</sup>. In the course of analysis of the structure of the major histocompatibility complex of the chicken, we found 2 exceptional birds among 1206 (CB×CC)F<sub>1</sub>×WB hybrids, 1 of which (cock 744) had a crossing over in the B complex<sup>6,7</sup>. The other cock (No. 2349) was further analyzed, and when its erythrocytes were used for absorption of specific anti-B1 (anti-CB) and anti-B2 (anti-CC) sera, both were completely absorbed. Using appropriate anti-B1 and anti-B2 sera, we detected 2 types of erythrocytes in this chicken and found that approximately 40% of the erythrocytes were agglutinated by anti-B1 and 60% by anti-B2 serum. The proportion changed within only ±5% in each of 5 measurements performed over 2 years. The erythrocyte chimaerism was confirmed when erythrocytes from cock 2349 were used for immunization. The recipients of the B<sup>1</sup>/B<sup>10</sup> genotypes formed anti-B2 or anti-B1 antibodies which did not differ in specificity from antisera prepared by immunization with erythrocytes from the B<sup>1</sup>/B<sup>1</sup> (CB) and B<sup>2</sup>/B<sup>2</sup> (CC) donors.

Transplantation tests showed that skin grafts from congenic lines CB and CC (differing from one another only at the B complex) and the inbred line WB survived in cock 2349, whereas its sibs accepted a CB or CC and WB graft, depending on their genotypes (table 1). To exclude the nonspecific takes of the grafts, the cock 2349 was grafted with skin from another inbred line IC which was rejected within 20 days. When skin grafts from cock 2349 were applied to CB×WB and CC×WB chickens, they survived permanently. The take of the grafts transferred from allophenic animals to the parental lines (in our hands, to CB×WB and CC×WB chickens) is also known in mice<sup>8</sup>. No splenomegaly occurred when peripheral blood lymphocytes from cock 2349 were injected into embryos of the genotypes B<sup>1</sup>/B<sup>10</sup> and B<sup>2</sup>/B<sup>10</sup>, whereas the same lymphocytes produced a 10-fold spleen weight enlargement in B<sup>13</sup>/B<sup>10</sup> embryos.

Furthermore, the progeny from cock 2349 mated to hens originating from inbred CB, CC, and WB lines were analyzed. The results of serological analysis are shown in table 2. Of the 84 chickens obtained from all the matings, 27 inherited antigen B1, 16 the antigen B2 and 41 the antigen B10 from cock 2349. 3 types of sperm cells were involved in all the matings. This is best illustrated in the cross of cock 2349 to the WB hen, where 6 offspring inherited antigen B10, 4 the antigen B1 and 1 the antigen B2. Erythrocytes with B1 and B2 antigens from birds from this mating were used for absorption of anti-B1 and anti-B2 sera which were produced by reciprocal immunization between congenic CB and CC lines. We found that antigens B1 and B2 inherited from cock 2349 did not differ from the original antigens.

Karyological analysis<sup>9</sup> of feather follicle cells and peripheral white blood cells from cock 2349 and of bone marrow cells from its progeny revealed the diploid number of macrochromosomes, but accurate counts of microchromosomes could not be made.

Among the various possible mechanisms of chimaera formation<sup>10,11</sup>, the following ones may be considered in re-

spect of the results obtained: a) Fusion of 2 blastoderms with both germ lines of the same genetic sex<sup>12</sup>. 2 or more blastoderms on the surface of the yolk mass have been reported by a number of investigators (for a review see Romanoff and Romanoff<sup>13</sup>). When these blastoderms are simultaneously fertilized, 2 embryos arise which may fuse and initiate formation of a chimaera during common embryonic development. b) Simultaneous fertilization of the nonextruded 2nd polar body and the oocyte. The resultant 2 separate zygotes may give rise to a chimaera during common embryonic development. We are not able to determine from the results obtained which of the 2 mechanisms would account for the chimaeric chicken observed.

Another mechanism has been considered by Oleson and Briles<sup>14</sup>. A spontaneous erythrocyte chimaeric chicken reported by them was an intersex which was apparently diploid, and appeared to have resulted from simultaneous fertilization of the regular gamete and the 1st polar body. These cells differ in their heterochromosomes, and after fertilization, the resultant chimaera will obviously consist of 2 coexisting cell populations with male and female chromosomes. The chimaeric chicken obtained in our experiments was a normal fertile cock which regularly formed 3 types of sperm cells. It was therefore unlikely that the above mechanism of chimaera formation would be involved in this case.

Table 1. Skin graft survival in cock 2349 and its sibs

Recipients	Survival of skin grafts from donors				2349
	CB	CC	WB	IC	
2349	+	+	+	—	NT
B <sup>1</sup> /B <sup>10</sup>	+	—	+	NT	+
B <sup>2</sup> /B <sup>10</sup>	—	+	+	NT	+

+, Skin grafts survived throughout the observation time (6 months–3 years); —, skin grafts were rejected within 20 days of transplantation; NT, not tested.

Table 2. Serological analysis of cock 2349 and its offspring

Hens mated to cock 2349 and their genotypes at the B complex	No. of offspring	Reactions with serum <sup>x</sup>		
		anti-B1	anti-B2	anti-B10
CB (B <sup>1</sup> /B <sup>1</sup> )	17	+	—	+
	9	+	+	—
	12	+	—	—
CC (B <sup>2</sup> /B <sup>2</sup> )	18	—	+	+
	11	+	+	—
	6	—	+	—
WB (B <sup>10</sup> /B <sup>10</sup> )	6	—	—	+
	4	+	—	+
	1	—	+	+
Cock 2349		+	+	+

<sup>x</sup> Alloimmune sera were specific. The designation of the blood group (B) system conforms to international nomenclature, allele designations are those currently used in our laboratory. Scoring of the reactions; +, positive reaction of erythrocytes with the corresponding serum; —, negative reaction.

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### On genetic variability in a population of the widespread gecko *Hemidactylus brooki*

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**Summary.** An electrophoretic investigation of 22 gene loci in a local sample of a widespread and ecologically extremely versatile gecko species has shown a low degree of heterozygosity, as is average in lizards and other vertebrates.

Of the reptilian species whose genic variation has been tested through gel electrophoresis of proteins<sup>1-3</sup>, none belong to the tropical family Gekkonidae, the largest lizard family. 3 species of the most prosperous genus of this family, *Hemidactylus brooki*, *H. mabouia* and *H. turcicus*, are of special interest in the comparison of genetic variability with adaptive strategies between ecological generalists and ecological specialists. Largely through human agency, the 3 *Hemidactylus* species have spread all around

the world, and they are to be found in the widest range of habitats from forest to downtown Cairo or São Paulo, from upland savanna to sea-level cliffs, from higher leaves in palm trees to under garbage heaps, etc. No other reptiles have such an ubiquitous distribution, including hundreds of other gekkonid species.

The opportunity having arisen to investigate a sample of 16 individuals (8 adult ♂, 5 adult ♀, 3 young) of *H. brooki* from the coastal town of Port-Gentil, Gabon<sup>4</sup>, we have

Table 1. Proteins and their encoding loci examined in *Hemidactylus brooki*. Enzyme commission numbers are in parentheses. Buffer systems as in Selander *et al.*<sup>5</sup>

Proteins	Loci	Active in	With the buffer systems
Lactate dehydrogenases (1.1.1.28)	<i>Ldh-A</i> <i>Ldh-B</i> * <i>Ldh-C</i>	Liver, kidney, heart, plasma, testis Liver, kidney Testis	Continuous Tris-citrate I Continuous Tris-citrate I Continuous Tris-citrate I
Malate dehydrogenases (1.1.1.37)	<i>Mdh-1</i> <i>Mdh-2</i>	Liver Liver, kidney	Continuous Tris-citrate II Continuous Tris-citrate II
Isocitrate dehydrogenases (1.1.1.42)	<i>Idh-1</i> * <i>Idh-2</i>	Liver, kidney Kidney, heart	Continuous Tris-citrate I Continuous Tris-citrate I
$\alpha$ -Glycerophosphate dehydrogenase (1.1.1.8)	<i>a-Gpd</i>	Liver, kidney	Tris-maleate
Phosphoglucomutase (2.7.5.1)	<i>Pgm</i>	Liver, kidney, erythrocytes	Tris-maleate
Phosphoglucisomerase (5.3.1.9)	<i>Pgi</i>	Liver, kidney	Tris-maleate
Glutamate oxaloacetate transaminases (2.6.1.1)	<i>Got-1</i> <i>Got-2</i>	Liver, kidney, heart, testis Liver, kidney, heart, testis	Continuous Tris-citrate II Continuous Tris-citrate II
Indophenol oxidase	<i>Ipo</i>	Liver, kidney	Continuous Tris-citrate II
Esterases (3.1.1.2)	<i>Est-1</i> <i>Est-2</i> <i>Est-3</i> <i>Est-4</i> <i>Est-5</i>	Kidney Liver, kidney, heart, plasma, erythrocytes Liver, kidney Liver, kidney Plasma, erythrocytes	Discontinuous Tris-citrate Discontinuous Tris-citrate Discontinuous Tris-citrate Discontinuous Tris-citrate Discontinuous Tris-citrate
Albumine	<i>Alb</i>	Plasma	Discontinuous Tris-citrate
General proteins	<i>Pt-1</i> <i>Pt-2</i> <i>Pt-3</i>	Plasma Plasma Plasma	Discontinuous Tris-citrate Discontinuous Tris-citrate Discontinuous Tris-citrate

Asterisks designate loci not included in heterozygosity and polymorphism computations (see text).