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Abstract: Our investigations into the causes of the cheetah's decline began in 1981, after Frank Brand, director of the National Zoological Gardens of South Africa, invited us, as representatives of the U.S. National Zoological Park, to cooperate in studying a seemingly narrow problem: why was it difficult to breed cheetahs in captivity? The analysis of 40 semen samples from cheetahs of Namibia and South Africa revealed that the concentration, motility and quality of sperm was significantly lower than usually seen in domestic cats. Two electrophoresis analysis of 52 and 155 proteins found no polymorphism in the first experiment and a frequency of only 3 percent of polymorphism in the second one. These results were confirmed by additional evidences. First, the skulls of the cheetah displayed a higher level of asymmetry than the skulls of three other cat species. Second, all skin graft exchanged between cheetahs individuals were accepted and were indistinguishable from the autographs of the 10-to-12-day period, indicating the monomorphism at the Major Histocompatibility Complex. The cheetah's genetic uniformity is certainly dangerous for the species, but we think it should not be interpreted as a death sentence.

The Cheetah in Genetic Peril

The world's fastest land animal is in a race for continued survival. An ancient population bottleneck has resulted in genetic uniformity and has made the species extremely vulnerable to ecological change

by Stephen J. O'Brien, David E. Wildt and Mitchell Bush

The cheetah, a virtual running machine, is a model of aerodynamic engineering. Its skull is small and lightweight and its limbs are long and slender, not unlike a greyhound's. Its heart, vascular system, lungs and adrenal glands are all enlarged, enhancing the animal's ability to accelerate and navigate during a high-speed chase. In addition the cheetah's claws are semiretractile: they are always extended like cleats, in contrast to the claws of other species in the cat family (*Felidae*), which are normally withdrawn and covered by a protective sheath. These various adaptations have made the cheetah a particularly effective hunter on the flat, open savannas of central and southern Africa, where it has a higher rate of successful kills than even the lion. After stalking its prey the cheetah launches a high-speed chase (often clocked at up to 70 miles per hour), pushes over or trips its winded victim and swiftly kills the prey by strangulation in its strong feline jaws.

In spite of the cheetah's impressive skill as a runner and hunter, the species seems to be heading for extinction. Where once the present-day species (*Acinonyx jubatus*) spanned the globe, now the cheetah's range is limited to a few small pockets in Africa. In all the world there are probably no more than 20,000 cheetahs.

How is one to explain the cheetah's march to extinction? Our investigations of the past five years suggest that the species has somehow lost its genetic variation. As a result of intensive inbreeding generations ago, each cheetah appears to be nearly identical with every other cheetah. Ever since Charles Darwin wrote *On the Origin of Species* a century ago it has been evident that genetic uniformity would hamper the ability of a species to adapt to such ecological perturbations as temperature shifts, drought, glaciation or the ascendance of new viruses or bacteria. Darwin's law of natural selection predicts that individuals well adapted to an environment will leave more offspring than less well-adapted individuals will. When a species has little genetic variety, its ranks are unlikely to contain many members whose genetically determined traits are well suited to withstand ecological change; the species competes poorly for survival under changed conditions and may die out.

As Darwin might have predicted, inbreeding has left the cheetah with traits that are as maladaptive as its lithe construction is adaptive. The species is vulnerable to disease and has an infant mortality rate that is estimated to be as high as 70 percent in some game preserves. In addition, although the cheetah is the world's fastest mammal, it can only run a few hundred yards. Having increased its respiratory rate from 60 to 150 breaths per minute during a typical chase, the animal often collapses for half an hour to regain its strength; during that time it can be attacked or, at the least, lose its bounty. Indeed, even when they are not exhausted, cheetahs are rather timid as defenders of their catch: fully 50 percent of their kills are snatched by more aggressive lions, leopards and hyenas.

Our investigation into the causes of the cheetah's decline began in 1981, after Frank Brand, director of the National Zoological Gardens of South Africa, invited us, as representatives of the U.S. National Zoological Park, to cooperate in studying a seemingly narrow problem: Why was it difficult to breed cheetahs in captivity? The early results of this cooperative effort yielded the first evidence that the cheetah might have a diminished gene pool and stimulated us to explore the cheetah's genetic status in detail.

Brand's problem was clear enough. In 1971 the Zoological Gardens had founded a cheetah-breeding program at the De Wildt Cheetah Breeding and Research Center, a compound near Pretoria. Ten years later the center led the world in numbers of offspring but was still frustrated by the animals' low fecundity and high infant mortality rate (37 percent).

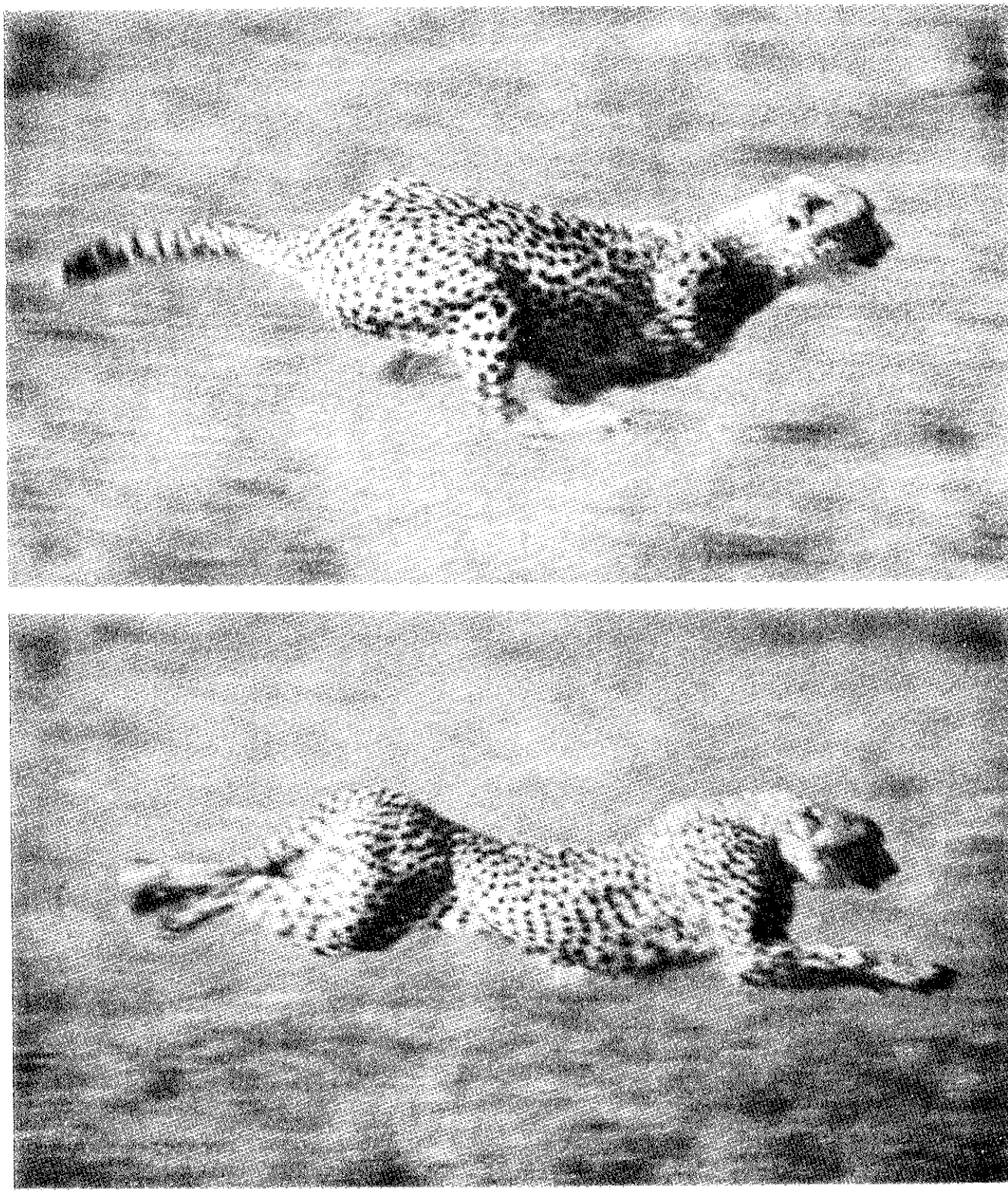
Indeed, the cheetah had a history of failure to breed in captivity dating back at least to the time of Akbar the Great, a 16th-century ruler in India. (Akbar, who had 1,000 cheetahs, was one of a long line of regal potentates on three continents who kept cheetahs as hunters and status symbols.) According to chronicles written by Akbar's son, the ruler had resorted to extreme efforts to promote breeding, including giving his regal specimens the run of the palace gardens. Even so, only a single litter was ever produced—and it was the sole recorded litter born to captive cheetahs until cubs were born at the Philadelphia Zoo in 1956.

Since 1956 a mere handful of breeding programs have been successful, and only from 10 to 15 percent of the sexually mature cheetahs caught in the wild have reproduced in captivity. Such low fecundity is often a consequence of unsuccessful mating attempts, but even after successful matings the cheetah has a low conception rate compared with that of other zoo-bred species, and about 30 percent of the cubs born in captivity die before the age of six months.

When we arrived at De Wildt to begin our diagnostic workup, including semen analyses and a study of genetic variety, the center had about 80 cheetahs. The colony had been established from two distinct regions of southern Africa: Namibia (South-West Africa) and the northern region of the Transvaal Province of the Republic of South Africa. The regions are separated by the Kalahari Desert, a

distance of 1,500 kilometers, which led the curators to conclude that the animals were representatives of separate geographic subspecies.

Our first step was to collect and analyze 40 semen samples from 18 males. The ejaculates were very different from those of other species we had studied. The concentration of sperm was only a tenth as high as the concentration usually seen in domestic cats, and the motility of the sperm was also significantly lower. Even more striking, sperm quality was invariably poor. In the average ejaculate some 71 percent of the sperm were shaped abnormally, in sharp contrast to the average of 29 percent found in domestic cats. For example, the flagella, which propel the sperm, were often coiled or bent at right angles, and many of the



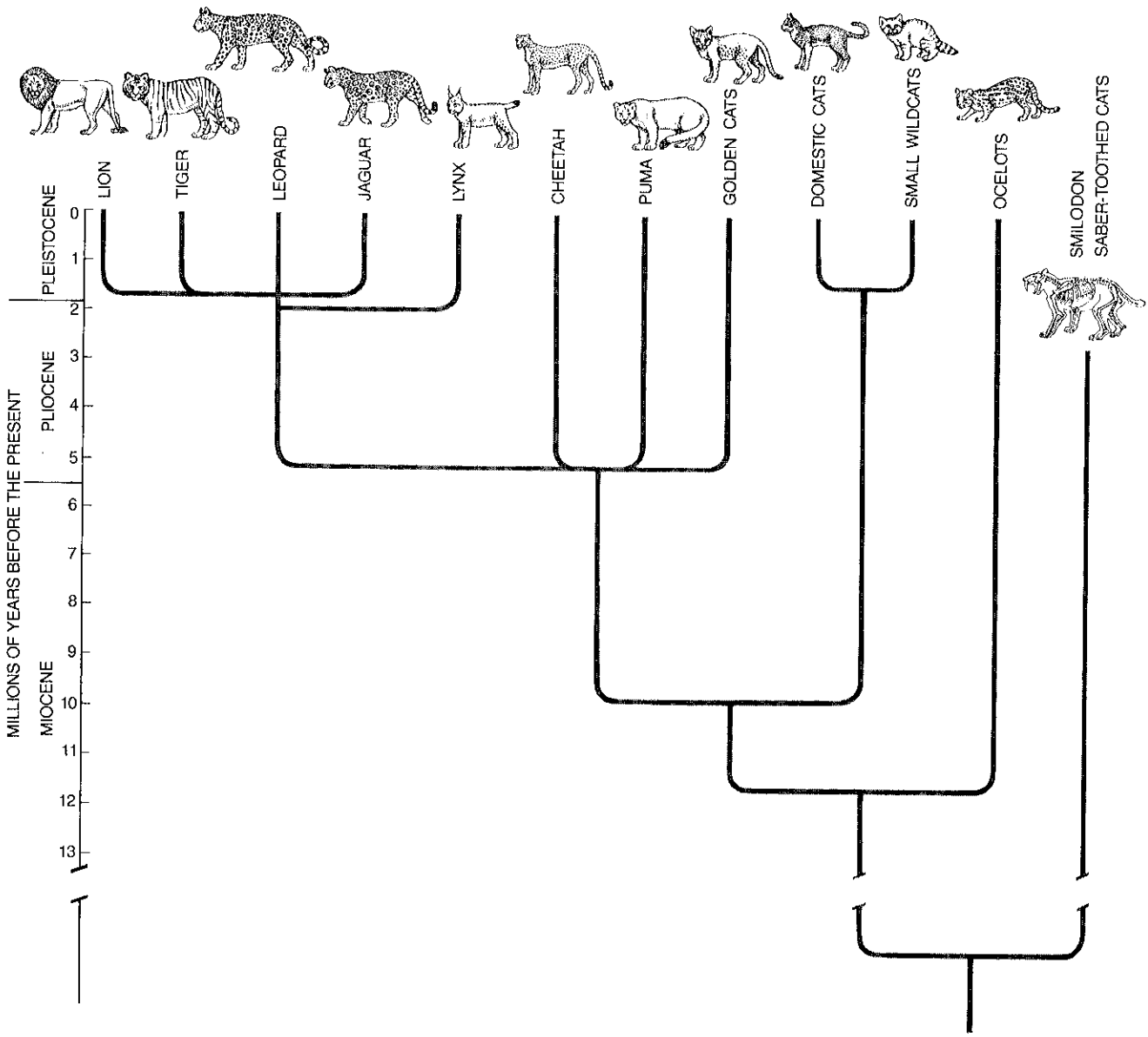
CHEETAH, the world's fastest sprinter, is shown in two of the positions it assumes while running at top speed. Often clocked at 70 miles per hour, the cheetah is airborne half of the time during a chase and can speed to 50 m.p.h. from a standing position in seconds.

sperm heads were either too large or too small.

The results of the semen studies seemed particularly significant because the vast majority of similar abnormalities (particularly at such a high incidence) in other species are associated with infertility. In the bull, for instance, the finding that 10 or 20 percent of the sperm are abnormal indicates that the animal is subfertile or even sterile. Furthermore, it seems clear that the quality of sperm morphology is under strict genetic control; increased morphological abnormalities often appear as a consequence of inbreeding of laboratory animals or livestock. The sperm data collected at De Wildt provided the first clue to the nature of the cheetah's plight.

To evaluate the extent of genetic variety in the De Wildt cheetahs we drew blood from 50 animals and arranged to have the blood-cell and plasma proteins—mostly enzymes—in the samples analyzed in the U.S. by gel electrophoresis, a standard procedure for the study of genetic variation. To make sense of the procedure one first needs to know a few basic facts about the mechanisms of genetic inheritance.

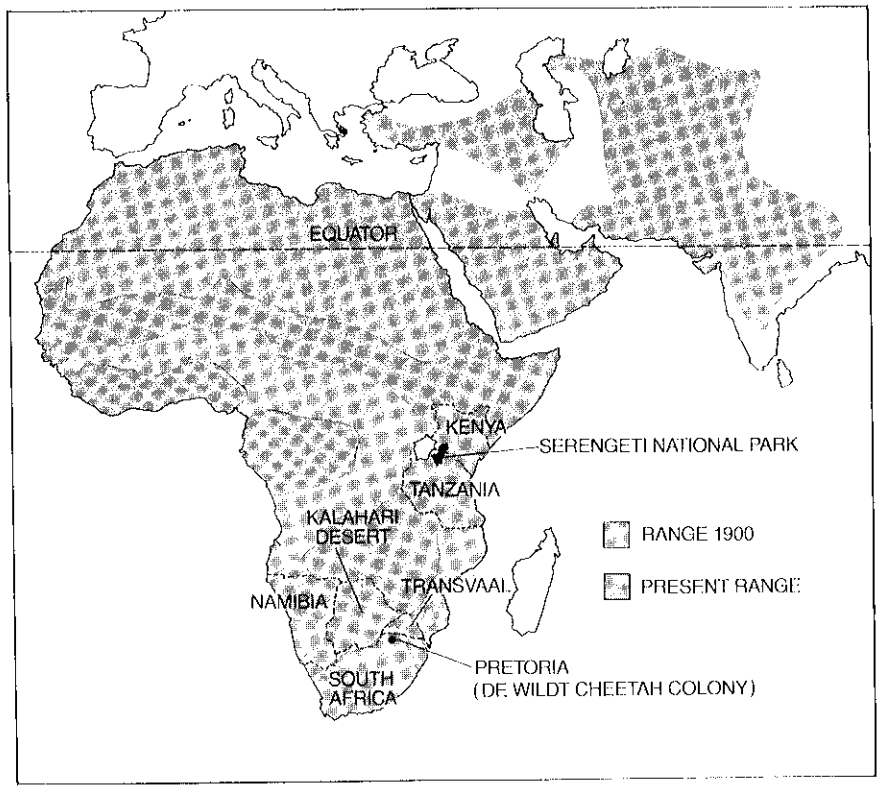
An animal receives a double set of chromosomes at conception, one set from the mother and an analogous set from the father. As a result each gene on a chromosome has an allele, or a mate, that resides on a paired chromosome and performs the same function as the first gene, such as directing the synthesis of a protein. Sometimes the alleles inherited from each parent at a given locus on the chromosome are homozygous, or identical, and sometimes they are heterozygous, or different. An individual that has heterozygous alleles for a given protein will produce two versions of that protein.



EVOLUTIONARY TREE of the cat family illustrates the relatedness of some of the 37 species that are in existence today. The fossil record shows that several families of saber-toothed cats came into being in the Miocene epoch (20 to 30 million years ago) and later died out, with one variety, Smilodon, persisting until 10,000 years ago. The modern felines began their radiation from other cats about 12 million years ago. The earliest branch led to the small and middle-size South American cats, including the ocelot and margay. The second branch began some eight to 10 million years ago and led to the domestic cat and its close relatives. The third branch began about four to six million years ago and led first to the pantherine lineage (including the golden cats, puma, cheetah, serval) and then to a split between the lynx and the modern big cats. This phylogeny is based on analyses of the protein albumin in modern cat species.

In carrying out a standard electrophoretic analysis of blood samples, workers isolate proteins from blood, place them on a gel and expose them to an electric field that causes the proteins to travel through the gel matrix. Proteins that are exactly alike (the products of identical genes) travel identical distances in the gel. Varieties of a given protein (the products of genes that encode proteins having an altered amino acid in the backbone) migrate to different positions. The proteins can be visualized with a specific stain, making it possible to compare the migration of the proteins and determine the degree of genetic variety within and between individuals [*see illustration on page 90*].

Our electrophoretic survey of the De Wildt cheetahs yielded a startling result. There was no variation in any of the 52 proteins examined; each cheetah was electrophoretically identical with every other cheetah. This was highly unusual. Similar electrophoretic surveys of genetic variation in more than 250 species over the preceding two decades had shown that between 10 and 60 percent of the genes in each species were polymorphic (had more than one electrophoretic form) and that between 1 and 36 percent of the genetic loci of the average individual were heterozygous. The genes of the cheetahs were monomorphic (had one form for each protein) and, it follows, were also homozygous at each protein locus. Such a striking level of genetic monomorphism is rare in natural populations. Like our semen analyses, it was reminiscent of the genetic homogeneity seen in purposely inbred species, such as certain laboratory mice.



RANGE OF CHEETAH is shown for today (*color*) and for the turn of the century (*shaded*). **In 1900 the range of the present-day cheetah, *Acinonyx jubatus*, extended through Africa, the Middle East and India. Now the cheetah is extinct as a free-ranging species everywhere except in central and southern Africa. Estimates of the number of cheetahs alive today vary from 2,000 to 25,000. Fossils of *A. jubatus* have been found in Europe, Asia, Africa and North America. Fossils of at least three other, extinct cheetah species have also been found: *A. pardinensis*, giant cheetahs that lived perhaps four million years ago in China, India and southern Europe; *A. intermedius*, a smaller species that ranged throughout Eurasia, and *A. trumani*, which had similarity to the puma and lived in the U.S.**

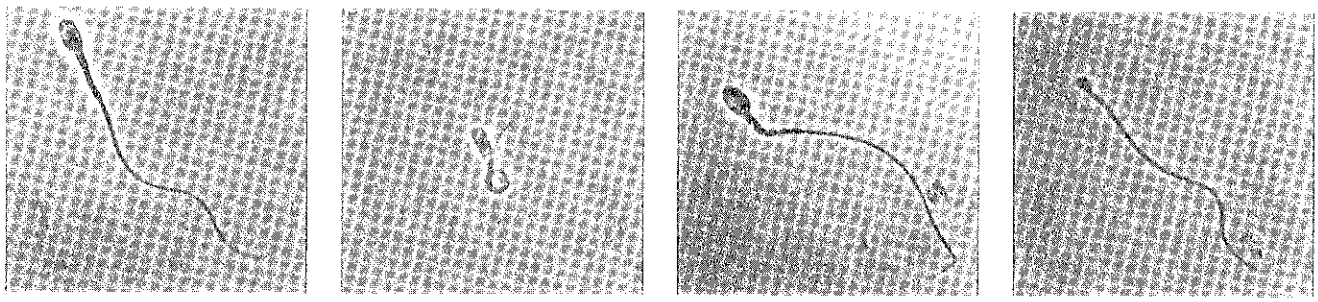
In our increasingly refined search for some indication of genetic variety, we took a somewhat different approach to protein analysis: two-dimensional gel electrophoresis. Like the technique employed in our enzyme surveys, this approach also analyzes protein migration in a gel, but it can evaluate hundreds of proteins simultaneously. We asked David Goldman and Carl R. Merril of the National Institute of Mental Health to examine proteins from the fibroblasts (connective-tissue cells) of six unrelated southern African cheetahs in U.S. and European zoos. Although these cheetahs did have a few variant proteins, the frequency of polymorphism among 155 proteins analyzed was found to be exceedingly low: 3 percent, or less than a third of what is usually seen in human populations that have been studied by the same technique.

The emerging profile of an inbred species in unusual genetic peril was corroborated by three additional studies, beginning with a comparison of cheetahs and other cats. Andrea Newman, a graduate student in the laboratory of one of us (O'Brien) at the National Cancer Institute (NCI), did a comprehensive electrophoretic survey of blood-cell enzymes from nine cat species housed in zoos and wildlife preserves throughout the world. Her results revealed moderate to high levels of genetic variety within each species studied, including some whose ranges overlap that of the African cheetah (the leopard, lion, serval and caracal) and some whose ranges do not (the tiger, ocelot, margay and domestic cat). In other words, the cheetahs we had studied were exceptional even among fellow felines in their diminished level of genetic variation.

The second set of data was obtained from an analysis of morphological traits in a group of African cheetah skulls collected in American museums. We estimated morphological variation on the basis of a measurement called fluctuating asymmetry: the extent to which features that are normally mirror images of each other, such as the left and right sides of the skull, are different sizes in a given individual. In a variety of species asymmetry has been found to increase in degree and incidence as a result of inbreeding, although just how the asymmetry comes about is not exactly clear.

Robert K. Wayne, a postdoctoral fellow in our laboratory at the NCI, evaluated fluctuating asymmetry in each of 33 museum-held skulls and compared the results with measurements from leopards, margays and ocelots, which are species we knew (from Newman's results) have abundant genetic variety. The skulls of the cheetahs did indeed display a higher level of asymmetry than the skulls of the other three cat species.

Our third -and most alarming- set of corroborative data came from a study designed to determine whether the cheetah displays any variability at the major histocompatibility complex (MHC). The MHC is a complex genetic locus in mammals that directs the synthesis of antigens on the surface



ABERRANT SPERM detected in animals at the De Wildt Cheetah Breeding and Research Center are shown, along with a normally shaped specimen (*far left*). Among the many striking abnormalities found were (*left to right*) coiling of the flagellum, bending of the midpiece and reduction in head size. The discovery that the cheetahs at the De Wildt center had a high incidence of abnormal sperm provided the first major clue that the cheetah's reproductive problems are physiological in nature and might have a genetic explanation.

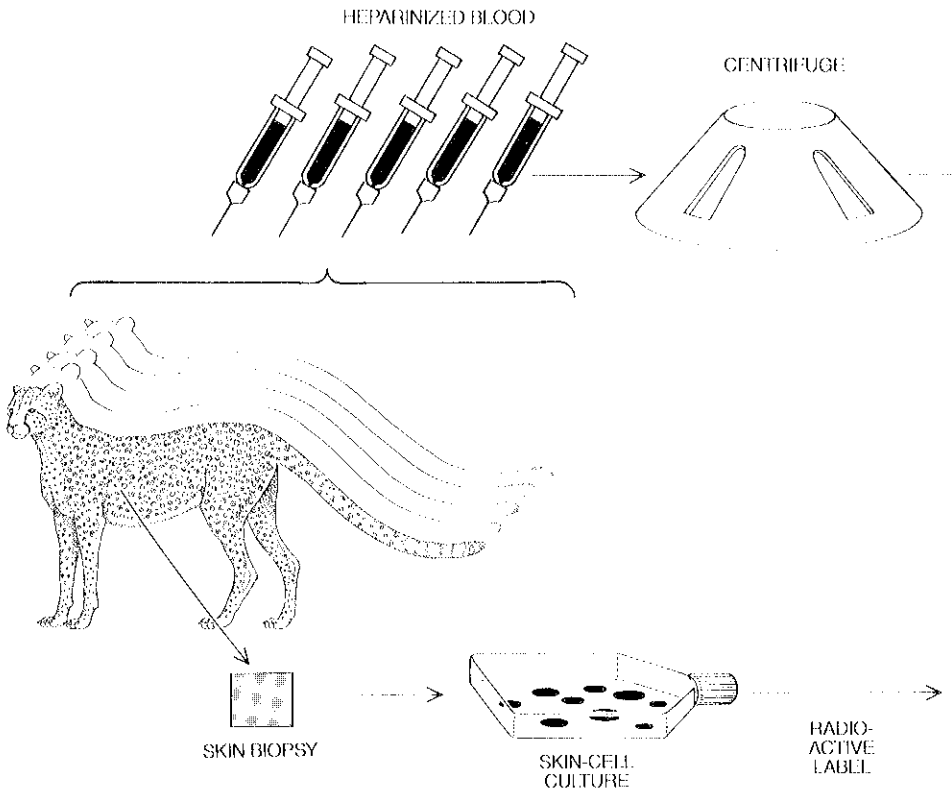
of most cells. The antigens communicate with *T* lymphocytes (circulating white blood cells) during an immune response to an infectious agent or to spontaneously arising aberrant cells, such as tumors. MHC antigens are also responsible for the immunological graft rejection that follows tissue or organ transplantation when the donor and the recipient have different MHC antigens. In the past 50 years the MHC system has been extensively studied in man and in the mouse, rat, dog, horse, chimpanzee and several other species.

The MHC is the most extensively polymorphic locus in mammals. The complex is really a chromosomal cluster having three functionally distinct groups of subloci, termed class I, II and III. In human beings the class I MHC sublocus alone is composed of three genes, each of which can be drawn from more than a dozen allelic varieties in the gene pool. As a result the number of possible combinations at the class I sublocus on one chromosome in humans is enormous (more than 12^3), and the chance of any two individuals having the same combination is slim (less than one in 10,000). Other mammalian species also display similar extreme polymorphism. The major exceptions are strains of inbred mice that are bred intentionally for homozygosity of their MHC genes and therefore accept skin grafts within but not between strains.

To assess the presence of polymorphism at the MHC in the cheetah, we exchanged skin grafts between the two members of seven pairs of cheetahs from southern Africa: six unrelated pairs and one sibling pair. A patch of skin was removed from each animal and cut in half. One piece was sutured into a graft bed in the paired recipient; it was an "allograft." The other piece was sutured into a graft bed in the animal from which it was removed; it was an "autograft." After the procedure the animals were examined and their grafts were cleaned twice a week for about eight weeks, during which time any signs of rejection were noted.

In several mammalian species, including domestic cats, the average survival time of skin grafts from an unrelated animal is some 10 to 12 days; rejection within this time is attributable to differences at the MHC. Slower, or chronic, rejection is the result of differences at other, minor histocompatibility loci in cases where there is identity at the MHC. Hence if a cheetah rejected an allograft within 10 or 12 days, we could conclude that the MHC genes of the two members of a pair were not identical.

Once again the cheetahs demonstrated their genetic uniformity. Remarkably, all allografts were accepted and indeed were indistinguishable from the autografts throughout the 10-



TWO ELECTROPHORETIC METHODS for measuring the extent of genetic variation in cheetahs are illustrated. In one method (*top*) fresh blood is treated with heparin to prevent clotting and allow different components (white cells, red cells and plasma) to be separated in a centrifuge. Soluble enzymes from the blood samples are then subjected to electrophoresis, that is, they are exposed to an electric field that causes them to migrate through a

to-12-day period. Three of the allografts did undergo slow rejection later, but several of the grafts persisted for at least 78 days, by which time they appeared to blend in with the recipient's own skin.

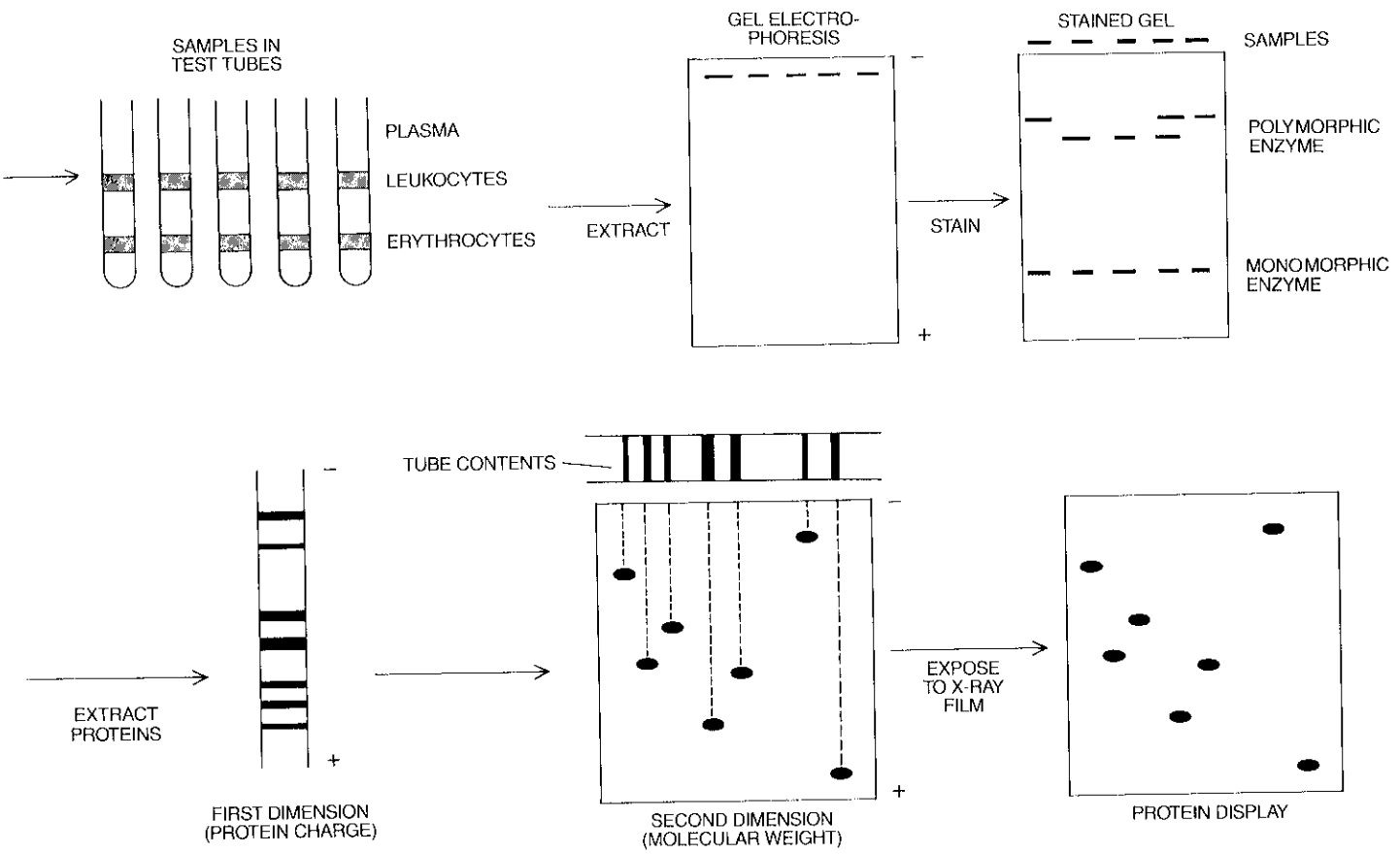
In order to be sure that the failure to reject the allografts was a result of identity at the MHC and not of a general failure of the cheetah's immune system to reject tissue grafts, we also sutured skin from a domestic cat into the graft bed of two of the cheetahs. In both cheetahs the graft from the cat underwent a classical acute rejection between days 10 and 12, whereas the autograft and the allograft continued to heal and grow. This indicated that the immune system of the cheetahs was indeed able to recognize antigens specified by foreign MHC genes. The cheetahs' immune system had simply not encountered any foreign antigens in the skin grafts from the purportedly unrelated cheetahs.

The cheetahs in these skin-graft experiments (and for that matter in all the studies we have described) demonstrate a level of genetic monomorphism that is unprecedented in any outbred mammalian species. Nevertheless, there is one possibility that must be considered before we can conclude definitively that cheetahs throughout Africa are virtual genetic twins and the products of inbreeding. The biochemical studies reported here all involved samples from cheetahs of southern Africa; most of the cheetah skulls that showed increased asymmetry (an indication of inbreeding) had been collected from eastern African populations by Theodore Roosevelt and his companions. There is a chance that the cheetahs in eastern and central Africa, although possibly inbred themselves, have gene pools somewhat different from those of the southern cheetahs. We are not optimistic that this is so, but it is a possibility we are exploring.

The fact that genetic uniformity poses a threat to the survival of a population or a species has been evident since Darwin formulated his theorem of natural selection. Genetic variation is the raw material for evolution; it is genetic heterogeneity on which natural selection operates in times of environmental or ecological change. The surprise, then, is that the monomorphic cheetah has persisted at all. It has not persisted without peril, however. Evolutionary theory predicts that a species with little genetic plasticity would be particularly vulnerable in a time of ecological perturbation, and a graphic demonstration that this is so for the cheetah occurred in 1982.

A pair of apparently healthy cheetahs arrived (on breeding loan) at the Wildlife Safari Park in Oregon, which had one of the most successful cheetah-breeding programs in the world. Within a few months the visiting cheetahs developed fever, diarrhea and jaundice, and they died a few weeks later. The cause of death was determined to be a viral infection called feline infectious peritonitis (FIP).

Although the disease spreads rampantly in colonies of domestic cats, FIP seldom kills more than 10 percent of infected animals. It was different for the cheetah. Within six months of the first two deaths at Safari Park, symptoms of FIP had developed in every cheetah in the park, and by the end of 1983 nearly half of the cheetahs had died of FIP-related disease. The epizo-



gel matrix, after which they are made visible by specific stains. Enzymes that are products of polymorphic genes migrate to different positions in the gel matrix, whereas enzymes that are products of monomorphic genes migrate to identical positions. In the second method (*bottom*) radioactively labeled proteins from skin cells that have been grown in culture are exposed to electric fields that separate them in two dimensions. The proteins are separated first on the basis of electric charge and then on the basis of molecular weight. Finally the gels are exposed to X-ray film, which reveals hundreds of proteins whose positions can be compared between cheetahs.

otic (an epidemic among animals) was the most extreme response to an FIP viral infection so far reported in any species. Since then we have learned of similar FIP epizootics at cheetah-breeding compounds in Ireland, Canada and Namibia.

One possible explanation for the Oregon epizootic was that the FIP virus was a particularly virulent strain and simply happened to strike the cheetahs first. This idea did not hold up. Several attempts to transmit FIP to domestic cats by inoculating them with virus collected from the cheetahs were unsuccessful. Moreover, 10 African lions in the Oregon compound remained free of symptoms after exposure to the virus. Such a lack of virulence in two feline groups indicated that the epizootic among the cheetahs was a result of cheetah vulnerability rather than of viral hypervirulence.

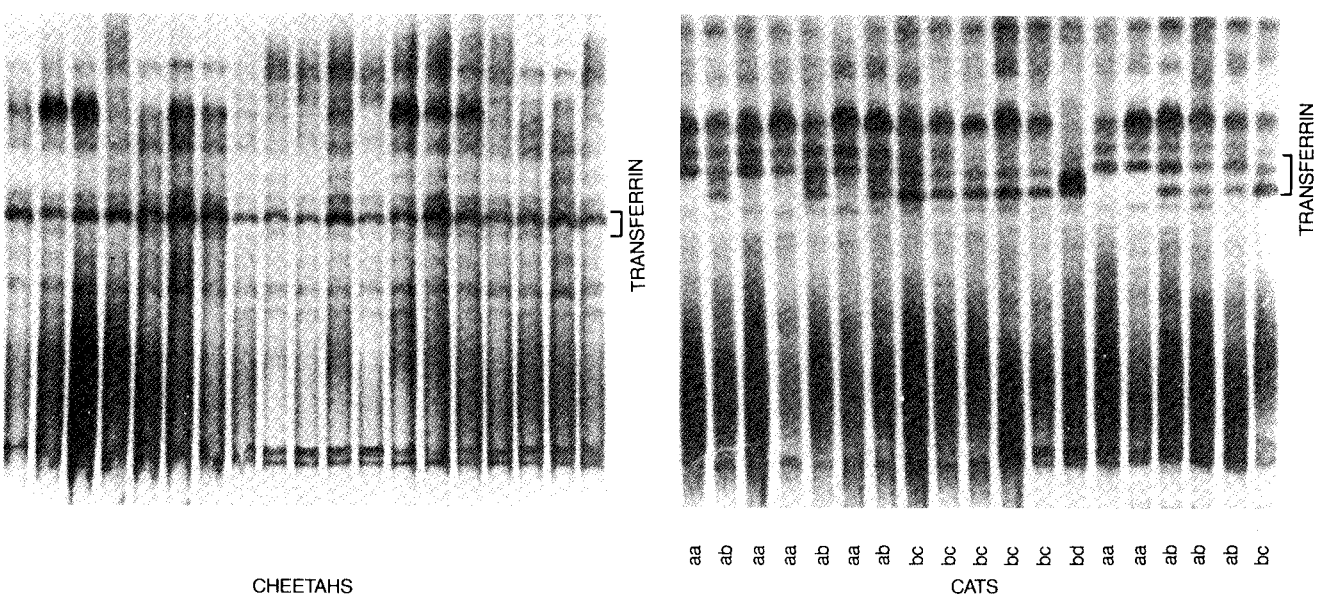
Monomorphism at the MHC, which would limit a species' repertory of defenses against a virus, is an attractive candidate for a biological explanation of the epizootic. In a monomorphic species one would expect widespread morbidity soon after a virus successfully overcomes one animal's defenses, because all the other animals would have much the same degree of susceptibility to the virus.

The event in Oregon could actually be explained by monomorphism of genes within either the class II or the class I MHC sublocus. The class II sublocus contains immune-response (IR) genes and is believed to contain viral-restriction genes also (although the two gene types may actually be the same entity). IR genes encode antigens that have been shown to elicit antibodies against synthetic antigens or viruses in laboratory animals. Viral-restriction genes have been shown in mice to determine the degree to which an organism can prevent the replication of viruses whose genetic material is RNA, and the FIP agent is an RNA virus. A population that is polymorphic at the MHC locus would be expected to have many varieties of IR and viral-restriction genes with the ability to confer protection against the FIP virus. On the other hand, a population that suddenly became monomorphic at the class II sublocus would be particularly vulnerable to a viral strain able to circumvent the immunological defenses that are controlled by the population's single, universally shared set of class II genes.

Monomorphism in the class I MHC sublocus would result in an epizootic by a different mechanism. Class I MHC genes encode antigens that must appear on the surface of a virus-infected cell before *T* lymphocytes can recognize viral antigens, which are also displayed on the cell surface, and kill the infected cell. Any virus able to alter itself in a way that prevents *T* lymphocytes from recognizing the class I MHC antigen on virus-infected cells would be effective against the host. In a polymorphic population any such adaptation to one host by a virus is unlikely to be effective against the immune system of a host having a different set of class I antigens. In a monomorphic population, however, any virus that adapts to one animal's immune-surveillance system would subsequently find every other system it encounters in the population to be identical to the first and therefore easy to get around.

It may be important to remember that exquisite strategies for abrogating immune surveillance are evolved by viruses in parallel with the defense systems of their hosts. Regardless of the exact explanation of the cheetah colony's reaction to the FIP virus, the episode in Oregon seems to emphasize the disadvantage of a genetically depauperate population in adapting to pathogens in its environment.

Having uncovered an array of evidence that the African cheetah is a genetically monomorphic species, we turned to the historical causes of the inbreeding that led to such monomorphism. Data relating to a single point in time in the dynamic evolution of a species do not give one much to go on. Nevertheless, we considered several hypotheses, each of which presumes that the ancestors of today's cheetah once had as much genetic variety as



ELECTROPHORETIC MIGRATION is shown for one protein, transferrin, in 19 plasma samples from cheetahs (*left*) and 19 samples from domestic cats (*right*). The transferrin that was synthesized by the cheetahs migrated to one position (*dark band across all samples*), indicating that all 19 cheetahs were monomorphic: they carried the identical transferrin gene. In contrast, the transferrin synthesized by the domestic cats migrated to several positions, revealing the presence of four different types of transferrin and indicating that the cat population was polymorphic: had more than one transferrin gene variant. (The letters at the bottom indicate the dual gene combinations expressed by each cat.) In the cheetah similar uniform results have been found for virtually all enzymes studied.

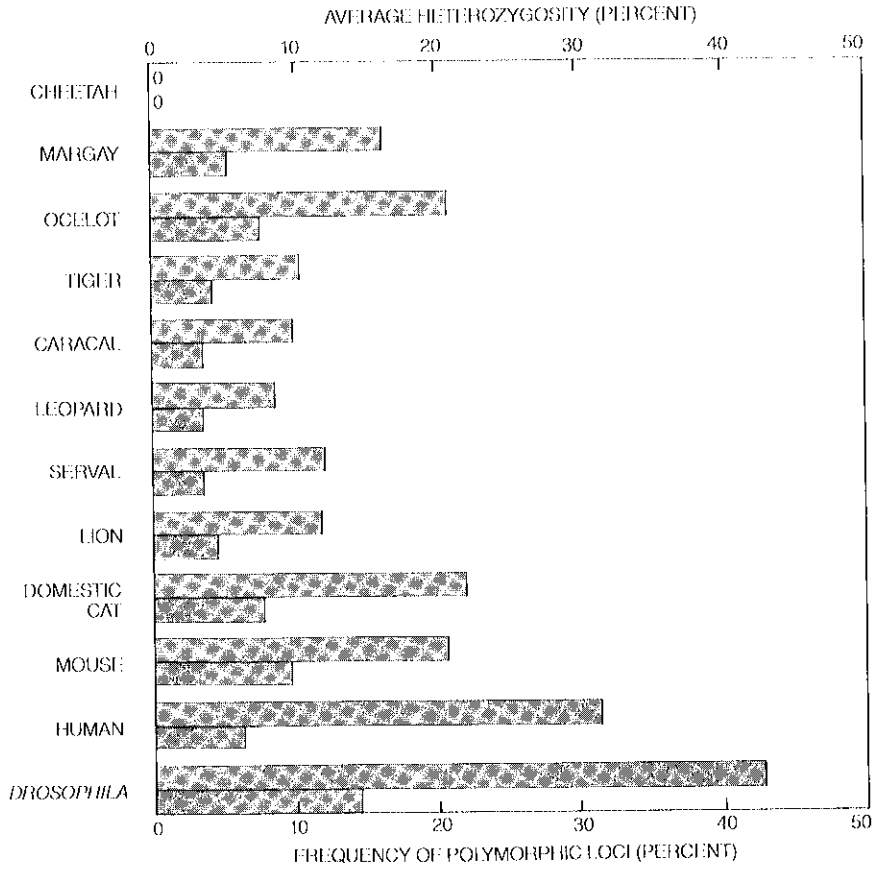
that currently enjoyed by other felines.

One hypothesis is that the cheetah is predisposed to incestuous mating, but that idea is not consistent with the findings of ethologists who have studied cheetahs of eastern and southern Africa. They find that cheetahs generally do not remain with their siblings after childhood. Male cheetahs are often territorial and occasionally nomadic, and unrelated females wander alone in and out of territories, mating with several males before moving on.

A second hypothesis is that the cheetah evolved to an adaptive optimum for a particular environmental niche and then gradually shed its variety during an extended period of niche stability. The idea is appealing to those who view the cheetah as a masterpiece of evolutionary construction, but various predictions based on this theory are not borne out by the facts. For example, a species ideally suited to a particular niche would be expected to have no closely related contemporaries, and yet the fossil record reveals that at least four species of *Acinonyx* have roamed the earth in the past few million years. One can only speculate about how many other species existed. An optimally adapted species would be expected to compete successfully with other species in its habitat, but the modern cheetah competes badly: predators always prevail in a showdown and cheetah young are often killed. A species ideally suited to its niche should have gradually shed its deleterious recessive alleles and acquired homozygosity only for highly adaptive traits, but the cheetah still has plenty of bad genes, for example those controlling sperm morphology.

Our third hypothesis, and the most plausible, is that at some point in the past the species went through an extreme population bottleneck: a severe population reduction. This was followed by inbreeding, which diminished the gene pool by the chance loss of alleles.

How extreme would a bottleneck have to be to produce a population with zero percent enzyme variation and with identity at the MHC? What caused the bottleneck in the cheetah populations and when did it occur? Theory and practice demonstrate that a population that passes through a bottleneck of a mere seven individuals can still retain about 95 percent of its original genetic variation; it can retain that variety if the survivors expand their numbers quickly and geometrically. (Slow expansion in a small population increases the likelihood that different gene types will disappear.) We therefore suspect that at least once and perhaps several times in the past the cheetah's forerunner populations must have dropped to a very few individuals, escaping extinction by a whisker; it is also possible that the surviving cheetahs never managed to expand their numbers rapidly. Just why the cheetah population would have dwindled so severely is anyone's guess. The possibilities range from climatic catastrophe to viral or bacterial plagues to destruction of the habitat or outright killing by humans.



GENETIC VARIATION in 12 species, including nine felines, is compared based on electrophoretic surveys of enzymes. ("Heterozygous" loci are positions on chromosome pairs where homologous genes differ from each other.) Of the more than 250 species that have been studied by population geneticists, the cheetah has the least amount of genetic variety.

The timing of the first bottleneck, like the degree and cause, is difficult to determine. A prime candidate is the time between 10,000 and 12,000 years ago, at the end of the geologic epoch known as the late Pleistocene. Before this there were many species of *Acinonyx;* the modern species, *A. jubatus,* had a worldwide distribution. Then something caused a massive extinction of mammalian species, particularly ones in North and South America, destroying 75 percent of those species. Many large carnivores succumbed, including cheetah species other than *A. jubatus,* and the range of the cheetah eventually became limited to its present one in certain parts of Africa. Whether the Pleistocene's environmental catastrophe did in fact cause a bottleneck in the population of *A. jubatus* is not at all certain. As likely as the notion seems, we cannot exclude the possibility that a more recent crisis is responsible for the cheetah's vulnerability today, and we hope our ongoing studies will provide more insight into the animal's past history.

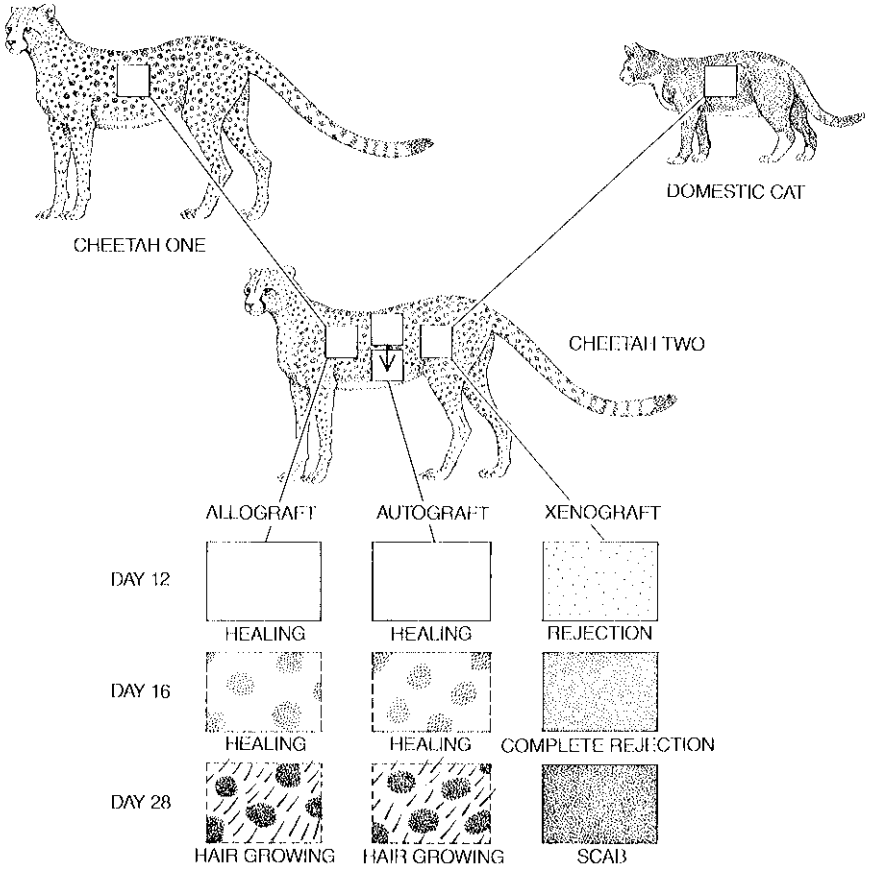
Our findings have revealed much about the past and present status of the cheetah, but an important component that must also be considered is the prospect for the future. The cheetah's genetic uniformity is certainly dangerous for the species, but we think it should not be interpreted as a death sentence. There are a number of reasons. If the proposed bottleneck did occur as long ago as the Pleistocene, then natural selection, which ensures that individuals with seriously maladaptive traits do not survive, has surely eliminated the most dramatically deleterious genes by now.

Several other animal species have gone through serious population bottle-

necks and seem to be recovering, and the cheetah may have the same good fortune. The northern elephant seal is an encouraging example. At the turn of the century the population was reduced to about 20 animals. Yet after (and perhaps because of) the passage of protective legislation in 1922 and later, the seal population off the coast of Mexico and California grew. Today the number of seals in the area reaches into the tens of thousands.

Finally, as we mentioned above, there is the possibility that the cheetah subspecies in eastern Africa is genetically different from the subspecies in southern Africa. If that is the case, the eastern African cheetahs might be introduced into captive-breeding programs, which with few exceptions have largely involved cheetahs from southern Africa. Even if the two groups seem, on the basis of our limited methods, to be genetically similar, one might still want to breed them together. This has been attempted successfully at the Whipsnade Zoo, outside London. Cheetahs that have mated there have produced litters reported to have a low infant mortality rate.

Human beings have been "civilizing" the wilderness for centuries, and the cost has too often been the demotion of evolution's most charismatic creations to endangered or threatened status. In the meantime much has been learned from the study of animals about human biology and evolution. We hope the future will see such knowledge applied to saving the world's threatened and endangered species. Perhaps science can even help the world's fastest mammal to win its race for survival.



PROOF OF CHEETAH'S MONOMORPHISM at the major histocompatibility complex (MHC) is depicted highly schematically. Three types of skin grafts were surgically transplanted onto cheetahs: allografts (from unrelated animals of the same species), autografts (from self) and xenografts (from another species). Both the allografts and the autografts were readily accepted and by day 28 after grafting were developing cheetahlike spots and growing hair. The xenografts, from a domestic cat, were rejected by day 12 after grafting. MHC genes determine whether a graft will be accepted; unless immunosuppressive drugs are given, a graft normally survives only if the donor has the same MHC genes as the recipient. Therefore the acceptance of the allografts by the cheetahs in the study could be explained in two ways: either their immune system was somehow suppressed or they had identical MHC genes. Rejection of the xenografts indicated that the cheetahs' immune systems were operating adequately, and so the animals must have had identical genes at the MHC.