Chromosomal Polymorphism in Sand Gazelles (Gazella subgutturosa marica)

M. Vassart, A. Greth, V. Durand, and E. P. Cribiu

A total of 84 Gazella subgutturosa from three captive populations (two in Saudi Arabia, one in Qatar) were karyotyped. The number of chromosomes is 33, 32, or 31 for the males and 32, 31, or 30 for the females because of the X:autosome translocation that is common in the genus and a centric fusion between the two pairs of acrocentric chromosomes. The G- and R-banded karyotypes of gazelles translocated show that this fusion is the same as that previously reported for gazelles from Jordan. The precise origin of these populations is not known, but in every case the first animals are said to come from the wild in Saudi Arabia. This chromosomal translocation appears to be a populational polymorphism and not the result of hybridization between two different subspecies of G. subgutturosa.

Nine out of 12 species of gazelles (Honacki et al. 1982) are classified as vulnerable or endangered (IUCN 1988). The following three species of gazelles are considered native to Saudi Arabia (Thouless et al. 1991): the goitered gazelle, or sand gazelle, Gazella subgutturosa, known locally as “rheem”; the Arabian gazelle, or “idmi,” G. gazella; and the Saudi gazelle, or “afiri,” G. (dorcas) saudiya. The rheem gazelle found in Saudi Arabia is thought to be a subspecies (G. subgutturosa marica), different from the one found in Persia (G. subgutturosa subgutturosa) (Harrison and Bates 1991). The only two areas in Saudi Arabia where the sand gazelles are still resident are the Al Harrah and Al Khunfah reserves in the north. However, two captive populations also exist: Thumamah, managed by the King Khaled Wildlife Research Center (KKWRC), and Qassim. Cytogenetic studies are useful for conservation of the genus gazella, as most species have different chromosome numbers, hybrids are easily detected. Further, more karyotypes of these species display original features: for example, the X chromosomes are very large and are involved in an X-autosome translocation (Effron et al. 1976). In 1988 Kingswood and Kumamoto described a chromosomal polymorphism in a herd of G. subgutturosa from Jordan. They argued the possibility that their sand gazelles could be hybrids between sand gazelles G. subgutturosa marica and Persian gazelles G. s. subgutturosa. In Saudi Arabia, a preliminary analysis using conventional methods showed the presence of a chromosomal centric fusion in the first sand gazelles brought to the Mahasat reserve from Thumamah (Granjon et al. 1991). Further work was undertaken to determine the frequency of this Robertsonian translocation in different herds of sand gazelles in the Arabian peninsula and to describe the chromosomes by different banding techniques.

Material and Methods

We analyzed a total of 84 animals (42 females and 42 males) from three populations.

Origin of the Animals

The sand gazelles we studied came from Thumamah (Saudi Arabia), Qassim (Saudi Arabia), and Qatar. Sand gazelles from the Saudi populations were caught and translocated to the Mahasat as Said reserve (200 km east of Taif, 21°59' to 22°31' N, 49°27' to 42°12' E).

Thumamah collection (near Riyadh, 25°03' N to 46°45' E). The individuals we studied originated from animals caught in the wild in different regions of Saudi Arabia (precise locations unknown) between 1976 and 1982. At the KKWRC, about 200 sand gazelles were present in 1986, a number that has since nearly doubled. In 1990, we transported 24 sand gazelles to the Mahasat reserve, and in 1991, we transported 28. From these and their offspring, we karyotyped 58 gazelles.

Qassim collection (27°09' N to 43°29' E) at the northwest of Riyadh. The first origin of these sand gazelles is said to be the Rub Al Khali (“Empty Quarter” of Saudi Arabia). In 1991, we caught about 45 sand gazelles; we transported 22 to a prerelease enclosure in the Mahasat reserve. Of the gazelles caught, we karyotyped 22.

Qatar collection. The collection belongs to the Ministry of Agriculture and Munic-
Table 1. Number of gazelles for the different chromosome numbers in the three populations

<table>
<thead>
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<th>Population</th>
<th>Males w/2n =</th>
<th>Females w/2n =</th>
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<tr>
<td></td>
<td>33</td>
<td>32</td>
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<td>31</td>
<td>31</td>
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<td></td>
<td>30</td>
<td>30</td>
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<tr>
<td>Qassim</td>
<td>5</td>
<td>3</td>
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<td></td>
<td>8</td>
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<td>2</td>
<td>2</td>
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<tr>
<td>Thumanah</td>
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<td>5</td>
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<td>1</td>
<td>28</td>
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<tr>
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<td>6</td>
<td>6</td>
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<tr>
<td>Qatar</td>
<td>1</td>
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ipal Affairs and is located at Ras Ushairij, in the north of the country. The exact origin is not known, but the first animals are said to have come from Saudi Arabia. We karyotyped four animals.

Methods

We established karyotypes from lymphocyte and fibroblast cell cultures. For lymphocyte cultures about 10 ml of peripheral blood was collected aseptically by jugular punctation into heparinized sterile glass tubes. We distributed 10 blood drops (0.5 ml) into vessels containing 9.5 ml of HAM'S F12 nutritive medium supplemented with 20% fetal calf serum, antibiotics (100 UI), and concanavalin A (10 mg/ml). The culture was then incubated at 37°C for 72 h, and colcemid (final concentration 0.03 mg/L) was added 1 h before harvesting. We then treated the cells with a hypotonic solution of sodium citrate (0.85%) for 20 min at 37°C, fixed them with Carnoy's solution, spread them on precooled slides, and stained them in a 4% Giemsa solution. The best metaphases were photographed and karyotypes were prepared.

For fibroblast cultures, we performed a biopsy of subcutaneous conjunctive tissue or muscle. After disruption and digestion in a trypsin solution (2.5 g/L) the cells were grown in a CO₂ incubator as monolayer cultures in Falcon dishes (75 cm²) containing a medium similar to the one previously described for lymphocyte culture. Once the cultures were established, we grew cells in MEM supplemented with 10% fetal calf serum, antibiotics, and glutamine. Cultures were synchronized with a single thymidine block during S phase (Hayes et al. 1991; Viegas-Pequignot and Dutrillaux 1978) in order to increase the yield of metaphase and early metaphase cells. To induce R-banding, we added 5-bromo-2-deoxyuridine (BrdU Sigma 5002) to the medium at a final concentration of 10 or 20 g/ml. The cultures were incubated at 37°C until the number of mitotic round cells reached a maximum, about 8–9 h after BrdU addition, as for sheep and goat cultures (Hayes et al. 1991). To obtain RBG bands, we treated the cells according to the procedure described by Hayes et al. (1991) and performed Fluorochrome-Photolysis-Giemsa (FPG) staining as described by Viegas-Pequignot et al. (1989).

G-banded karyotypes were prepared using a modification of Seabright's (1971) method. Chromosomes were ordered as in Kingswood and Kumamoto (1988). We obtained C bands by the barium hydrox-

Figure 1. (A) RBG karyotype of male Gazella subgutturosa with 32 chromosomes; (B) RBG-banded X chromosomes of a female.
one X and two acrocentric Y: Y₁, which is the true Y, and Y₂, which is autosomal. The G- and R-banding patterns of the additional autosome (Y₂) and of the distal part of the long arm of the X chromosome appear identical (Figure 1). G-banding patterns of the X chromosomes have been compared with other gazelle species for which G-band karyotypes are published. Our results show that for G. subgutturosa the autosome involved in the translocation is the same one as in G. soemmerringi (Benirschke et al. 1984) and G. dama (Arroyo Nombela et al. 1990). The X p-arm is entirely darkly stained by the C-banding technique (Figure 2).

**Discussion**

As for the other species of gazelles, males have an additional chromosome compared to females because of the X-autosome translocation (Effron et al. 1976).

The RBG-banding technique delineates the autosomal region of the X chromosome. Using this technique it is possible to distinguish between early (active) and late-replicating (inactive) chromosomes and chromosomal regions. The late-replicating X chromosome appears uniformly pale and more elongate than the early-replicating one, except in the pseudoautosomal region (Figure 1), which escapes the inactivation.

In mammals X-autosome translocations have also been described in Gerbillidae (Viegas-Pequignot et al. 1982), marsupial species (Sharman 1961), and man and mouse (Searle 1962). In G. soemmerringi (Benirschke et al. 1984) and G. dama (Arroyo Nombela et al. 1990) an unusual sex-determining mechanism has been described: X₁X₂Y₁Y₂ for males and X₁X₂X₁X₂ for females, due to X-autosome and Y-autosome translocations.

Figure 2 shows that in G. subgutturosa the p-arm of the X is composed of constitutive heterochromatin, as was described by Furley et al. (1988) for G. benetti.

The Robertsonian translocation found in our three captive populations is the same as the one described by Kingswood and Kumamoto (1988) on G. s. marica from eastern Jordan. To date, the only G. s. subgutturosa karyotype described is the one with 31 chromosomes for the males and 30 for the females (Kingswood and Kumamoto 1988), leading Kingswood and Kumamoto (1988) to state that their results did not rule out the possibility of hybridization between two subspecies. The fact that the same translocation has been

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**Figure 2.** (A) GTG karyotype of male Gazella subgutturosa with 32 chromosomes; (B) C-banded X chromosomes of a female.
detected in three different populations inside the Arabian peninsula (plus the one from Jordan) leads us to suggest that this translocation occurs as a populational polymorphism. Furthermore, because recent investigation shows that the first sand gazelles from Qassim came from the Rub Al Khali, the translocation must have occurred in the wild in this part of the peninsula. Since this translocation has been observed in different populations of the peninsula, it could be of very ancient origin and could have appeared de novo in the wild before the capture. Distribution of frequencies for the different chromosome numbers was found to conform with Hardy-Weinberg equilibrium (nonsignificant chi-square values between observed and expected numbers of the different karyotypes) for the Thumamah population, as well as for the three populations pooled together. Our results could be explained by a chromosomal gradient over the whole range of G. s. marica and G. s. subgutturosa. If this is true, we would have G. s. marica with 33/32 chromosomes in the southwest of the Arabian peninsula (the most western part of the range of the species) and G. s. subgutturosa with only 31/30 chromosomes in the most eastern range of this subspecies. However, random drift in the captive populations with low effective numbers and weak selection pressure against the presence of the translocation make it difficult if not impossible to extrapolate the frequencies found in our samples to the situation in the wild.

Gazelles that are carriers of the translocation do not exhibit phenotypic alterations, as was already found (Granjon et al. 1991; Kingswood and Kumamoto 1988). All the females have horns (in G. s. subgutturosa most of the females do not have horns). Chromosomal polymorphism in Artiodactyle is increasingly being documented, even if descriptions of wild specimens are still scarce. Among the Antilopinae family, chromosomal polymorphism has been described in the impala (Aepyceros melampus) (Wallace 1977, 1980) and in the blackbuck Antilope cervicapra (Eftron et al. 1976). Benirschke et al. (1984) found 12 different karyotypes in Soemmerring’s gazelles because of different centric fusions, and Arroyo Nombela et al. (1990) have described a centric fusion in G. dama.

Finally, it seems important to identify precisely the chromosomes involved in the X-autosome translocation for each taxon. For example, inside the Antilopinae it is important to know if the X autosome of the blackbuck (Antilope cervicapra) has the same origin as in the gazelles. If it does, the translation would have appeared only once in the family.

Chromosomal polymorphism should be explored by a cytogenetic survey on wild populations whenever it is possible. For example, such a study should be undertaken in Iran on G. s. subgutturosa to check the chromosomal gradient hypothesis.

From the National Wildlife Research Center, National Commission for Wildlife Conservation and Development, Tali, Saudi Arabia (Vassart and Greth) and the Institut National de la Recherche Agronomique, Centre de Recherche de Jouy-en-Josas, Laboratoire de Cyto- genetique, Jouy-en-Josas, France (Durand and Cri- bin). This work was carried out under the patronage of His Royal Highness Prince Saud Al Faisal and Dr. Abuzinada, Secretary of the National Commission for Wildlife Conservation and Development. The authors thank H. Hayes for help in translation of the manuscript. Please address reprint requests to Dr. Vassart at the Ecole Veterinaire de Toulouse, 23 chemin des Capelles, 31076 Toulouse, France.

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References


