Genetic Variability in the Arabian Oryx (Oryx leucoryx)

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An electrophoretic survey of blood markers of the Arabian oryx (Oryx leucoryx) was undertaken in order to ascertain the genetic variability in a sample of 85 individuals, mainly from Saudi Arabian (Taif) and Jordanian herds. Three out of 18 loci were found to be polymorphic (P = 16.7%) and the mean heterozygosity (H = 0.052) appears to be relatively high with respect to the severe demographic bottlenecks expressed by the species since the 1960s. No genetic differentiation was found between Arabian and Jordanian samples considered. Consequences of these findings for the management of the Taif herd and for such procedures as pedigree determination are discussed, and an example of this latter application is given for a case of doubtful parentage.

Key words: electrophoresis, blood proteins, heterozygosity, captive-breeding management

INTRODUCTION

The Arabian oryx (Oryx leucoryx) disappeared in the wild during the 1970s due to habitat degradation and over-hunting [Abuzinada et al., 1988]. The World Herd was created in 1963 to save the species from extinction; 9 oryx were sent to the United States, 7 of which qualify as founders (3 wild-caught, 3 from Riyadh Zoo, and one from London Zoo [Homan, 1988]). In the 1970s and 1980s, a number of programs were started in the Middle East, with the goal of reintroducing this species to the wild [Stanley Price, 1989].

In 1986, HRH Prince Saud Al Faisal in Saudi Arabia established the National Commission for Wildlife Conservation and Development and the National Wildlife Research Center (NWRC) in Taif, one of the aims of which was the breeding of the Arabian oryx for reintroduction to the wild. Unfortunately, due to an outbreak of tuberculosis among the Taif herd, oryx from other sources had to be used for the first
release in Saudi Arabia (Mahazat As Said Reserve). Today, after various measures have been taken, the health of the Ta'if herd is continuing to improve. The first calves were recently born from hand-reared females and, being free of tuberculosis, will be released in the near future.

Although experimental demonstrations are still scarce, it is generally considered that heterozygosity and fitness are positively correlated [Allendorf and Leary, 1986; Templeton et al., 1987; Hedrick et al., 1986]. Thus, captive-breeding programs should aim to maintain as much heterozygosity and allelic diversity as possible, these two parameters providing good estimates of both the short-term and long-term survival and adaptation abilities of populations [Allendorf, 1986]. Knowing the genetic variability of captive managed populations of rare or endangered species is therefore of great importance, especially when considered for return to the wild.

The purpose of this study was to determine the genetic variability in the captive Arabian oryx herd in Ta'if, and in the first oryx nucleus recently released in the Mahazat As Said Reserve, by protein electrophoresis. For comparison, we also analysed a sample from the Jordanian herd of Schaumari Wildlife Reserve [Abu Jafar and Hays-Shanin, 1988], and a few individuals from captive herds of Bahrain and Qatar. The results are compared with those obtained on other artiodactyls and are discussed in light of the recent history of the Arabian oryx and in relation to its captive breeding and reintroduction in Saudi Arabia.

MATERIALS AND METHODS

Origin of the Animals

The NWRC herd originated from a herd of 56 oryx kept at the late King Khaled’s farm in Thumamah (Fig. 1). This herd was established in the 1970s, derived from captive herds in Qatar, Bahrain, and Saudi Arabia (Riyadh Zoo), as well as wild-caught founders. Precise origins and identity of these various founder stocks are not known, because of the lack of any written document. Since this population was not genetically managed, inbreeding and genetic drift probably occurred. Twenty-nine oryx from the American World Herd were added to the Thumamah herd in 1982. After the arrival of the oryx at the NWRC in 1986, a tuberculosis outbreak reduced the herd from 56 to 35 individuals. These survivors are the “founders” of the current herd (see Fig. 1).

The Mahazat As Said herd originated from 5 Jordanian animals and 8 from the San Diego Zoo. Two of the San Diego oryx were removed from the group before release, and are not included in the study although a calf from one remains in the herd. On the other hand, of the 6 offspring born during the pre-release period, 3 were conceived from two sires before shipment of the dams from the United States. Two of these calves are included in the study. Altogether, 17 oryx were reintroduced on March 1, 1990.

The herd at Schaumari (Jordan) derives from 8 animals from the U.S.A. and 3 animals from Qatar [Abu Jafar and Hays-Shanin, 1988]. The Qatari herd is descended from animals caught in late 1950s and early 1960s in the south of Rub Al Khali in Saudi Arabia and western Oman [Jones, 1988]. A chromosomal translocation recently found both in one of the two Qatari animals considered here and in the Schaumari herd [Cribiu et al., 1990] suggests that the former came from the same ancestral group as those which founded the Jordanian herd. The herd of Bahrain
(Al-Areen Wildlife Sanctuary) is divided into 2 unmixed groups. One originates from animals issued from Saudi Arabia in the seventies [Jones, 1988]. The origin of the second group is unclear and may be Qatar or Abu Dhabi (United Arab Emirates). The animals from Bahrain studied here are supposed to belong to the second group.

**Blood Samples**

Blood samples were taken from 61 oryx of the Taif herd (present population 104), 11 in the Mahazat As Said herd, 14 in the Jordanian herd (which today numbers about 95), 3 from Bahrain, and 2 from Qatar. Blood was collected by jugular puncture after anesthesia and was stored (between 1 and 10 hours) in heparinized tubes at 4°C until treatment. Saline solution was added before centrifugation to separate plasma from red cells. The red cells were then washed with saline before hypotonic shock with distilled water. The serum and red cell samples were kept at −30°C until
TABLE 1. Enzymes and buffer systems*

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Tissue</th>
<th>Loci</th>
<th>Buffer system</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspartate-aminotransferase</td>
<td>RBC</td>
<td>1</td>
<td>TME 6.9/TME 6.9</td>
</tr>
<tr>
<td>Diaphorase</td>
<td>RBC</td>
<td>2</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Esterase</td>
<td>RBC</td>
<td>2</td>
<td>TME 6.9/TME 6.9</td>
</tr>
<tr>
<td>Glyoxalase</td>
<td>RBC</td>
<td>1</td>
<td>TBE 8.6/TBE 8.6</td>
</tr>
<tr>
<td>Glucose phosphate isomerase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Lactate dehydrogenase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Malic-enzyme</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Malate dehydrogenase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Purine nucleoside phosphorylase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Phosphogluconate dehydrogenase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Superoxyde dismutase</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Hemoglobin</td>
<td>RBC</td>
<td>1</td>
<td>TC 6.4/TC 6.0</td>
</tr>
<tr>
<td>Albumin</td>
<td>Serum</td>
<td>1</td>
<td>LiOH 8.3/LiOH 8.1</td>
</tr>
<tr>
<td>Esterase</td>
<td>Serum</td>
<td>1</td>
<td>LiOH 8.3/LiOH 8.1</td>
</tr>
<tr>
<td>General protein</td>
<td>Serum</td>
<td>2</td>
<td>LiOH 8.3/LiOH 8.1</td>
</tr>
</tbody>
</table>

*See text and Pasteur et al. [1988] for buffer systems description; RBC = red blood cells.

Electrophoresis was performed. Storage duration at −30°C before experiments was highly variable (between 2 weeks and 12 months).

**Protein Electrophoresis**

Horizontal starch-gel electrophoresis was conducted according to the method of Pasteur et al. [1988], with starch concentration of 12%. Staining procedures followed those of Pasteur et al. [1988]. Loci and buffers utilized are listed in Table 1. Hemoglobin was scored as a single locus, as described in Ryder et al. [1981], although it is probably encoded by 3 loci. The percentage of polymorphic loci (P) and the mean number of alleles per locus (A) were calculated for the whole sample, whereas mean heterozygosities (H) were calculated from allele frequencies in the subsamples defined above, as well as in the whole sample. Also, Nei’s genetic distances between the Taif, Jordanian, and Mahazat As Said groups were computed.

**RESULTS**

Three out of 18 loci (Ldh, Sod, Hb) were found to be polymorphic (P = 16.7%). Photographs and interpretative diagrams of Sod and Ldh polymorphisms are presented in Figure 2. The mean number of alleles per locus was 1.17. Heterozygosity at each of the three variable loci was calculated (Table 2), and all three loci were found to conform with Hardy-Weinberg equilibrium (non-significant Chi-square values between observed and expected numbers of the different genotypes). Mean heterozygosities for the five defined groups and for the whole sample are presented in Table 2. The 85 Arabian oryx considered here had a mean overall locus heterozygosity of 0.052, with the ones of the Taif group, the Jordanian group, and the Mahazat reintroduced group being respectively 0.055, 0.049, and 0.033. Results concerning the samples from Qatar and Bahrain will not be discussed, due to small sample size.
Fig. 2. Photographs and interpretative diagrams of superoxide dismutase (SOD) and lactate dehydrogenase (LDH) polymorphism in Arabian oryx.

DISCUSSION

In her analysis of the genetic status of the Arabian oryx, Mace [1988], using pedigrees given by international studbooks, identified 18 founders and estimated by pedigree simulation that only a small proportion of each founder's genome was expected to have been lost, mainly because of the rapid increase in population size and a well-managed breeding program. The results presented here also reflect a relatively high variability, at least for the studied loci, with proportion of polymorphic loci, mean number of alleles per locus, and mean heterozygosity being, respectively, 16.7%, 1.17, and 0.052. Baccus et al. [1983] gave mean P values of 11.4, 13.2, and 12.8% for large grazing mammals, large mammals, and all mammals. Comparisons with different species of artiodactyls (Table 3) also show that polymorphism and heterozygosity of the Arabian oryx fit well within the range of known values. The same applies to A, also in the range of those given by Baccus et al. [1983] for large grazing mammals (between 1.00 and 1.52). However, all these findings would be worth verifying on a larger sample of loci.

As far as the NWRC Saudi herd is concerned, a possible explanation for this
TABLE 2. Allele frequencies and heterozygosities (H) at three polymorphic loci, and mean heterozygosities (H) at 18 loci for five subsamples and the whole sample of Arabian oryx*

<table>
<thead>
<tr>
<th>Locus Alleles</th>
<th>Taif (N = 61)</th>
<th>Jordan (N = 14)</th>
<th>Mahazat (N = 10)</th>
<th>Bahrain (N = 3)</th>
<th>Qatar (N = 2)</th>
<th>Overall (N = 85)</th>
</tr>
</thead>
<tbody>
<tr>
<td>80 SOD</td>
<td>0.221</td>
<td>0.214</td>
<td>0.200</td>
<td>0.333</td>
<td>0.500</td>
<td>0.229</td>
</tr>
<tr>
<td>100 SOD</td>
<td>0.779</td>
<td>0.786</td>
<td>0.800</td>
<td>0.667</td>
<td>0.500</td>
<td>0.771</td>
</tr>
<tr>
<td>80 LDH</td>
<td>0.180</td>
<td>0.179</td>
<td>0.050</td>
<td>0.333</td>
<td>0.500</td>
<td>0.182</td>
</tr>
<tr>
<td>100 LDH</td>
<td>0.820</td>
<td>0.821</td>
<td>0.950</td>
<td>0.667</td>
<td>0.500</td>
<td>0.818</td>
</tr>
<tr>
<td>100 Hb</td>
<td>0.784</td>
<td>0.857</td>
<td>0.900</td>
<td>1.000</td>
<td>1.000</td>
<td>0.823</td>
</tr>
<tr>
<td>130 Hb</td>
<td>0.316</td>
<td>0.143</td>
<td>0.100</td>
<td>0.000</td>
<td>0.000</td>
<td>0.177</td>
</tr>
</tbody>
</table>

*Five individuals are common to the Jordan and Mahazat samples.

Relatively high variability may be that the initial founders of the Taif herd had different origins. This could explain, for instance, a frequency of more than 17% for the fast allele of Hb, a higher value than the 6% recorded by Ryder et al. [1981] from 31 individuals in American zoos. In addition, demographic bottlenecks have been of modest duration (Fig. 1), and therefore probably did not have much impact on heterozygosity. Although impossible to be determined precisely, the loss of H was probably less than 20%, which approximately corresponds to the one observed after a bottleneck of 25 individuals during 10 generations [Allendorf, 1986]. The same hypothesis may be invoked for the Jordanian group, as described by Abu Jafar and Hays-Shanin [1988].

Allelic frequencies and heterozygosity comparisons between the different subsamples yield some interesting results (Table 2). The Taif and Jordanian groups show similar H values (0.055 and 0.049, respectively), and a small Nei’s genetic distance (D) of 0.003. The sample from the reintroduced group in Mahazat As Said displays a lower mean heterozygosity, mainly due to lower frequencies of the less frequent alleles in Ldh and Hb (see Table 2), and somewhat higher D with the Taif and Jordanian groups (0.012 and 0.007, respectively). These rather narrow genetic distances, however, reflect the recent common origin of most of the studied animals.

The results of this study can have practical implications for the management of the Taif captive-bred oryx herd. Several procedures should therefore be applied in order to preserve as much heterozygosity as possible, and also to help in pedigree analysis [Ryder et al., 1981; Hedrick et al., 1986]: 1) In the near future, the selection of sires should be based on electrophoretic patterns at the polymorphic loci as well as on pedigrees or morphological analysis [see Wayne et al., 1986] in order to limit inbreeding. 2) Large-scale electrophoretic screening of the different herds maintained in the Middle East (Jordan, Qatar, Bahrain, Oman) and elsewhere in the world should be carried out in order to allow selection of candidates for introduction to the NWRC herd or to the wild. This information will help to preserve or increase the level of heterozygosity [for computer simulation, see Lacy, 1987], and also the allelic diversity [Fuerst and Maruyama, 1986]. For the next reintroductions in Mahazat As Said,
TABLE 3. Number of individuals, number of loci, mean heterozygosities (H), and percentages of polymorphic loci (P) for various species of artiodactyls*

<table>
<thead>
<tr>
<th>Species</th>
<th>N ind.</th>
<th>N loci</th>
<th>H (%)</th>
<th>P (%)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>White-tailed deer (Odocoileus virginianus)</td>
<td>1571</td>
<td>26</td>
<td>9.7</td>
<td>42.3</td>
<td>Kennedy et al., 1987</td>
</tr>
<tr>
<td></td>
<td>753</td>
<td>19</td>
<td>7.4</td>
<td>35.8</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td></td>
<td>10-1549</td>
<td>19</td>
<td>3.6–7.8</td>
<td>10.5–36.8</td>
<td>Smith et al., 1986</td>
</tr>
<tr>
<td>Red brocket (Mazama americana)</td>
<td>52</td>
<td>19</td>
<td>7.0</td>
<td>57.9</td>
<td>Smith et al., 1986</td>
</tr>
<tr>
<td>Brown brocket (Mazama gonazobira)</td>
<td>1</td>
<td>19</td>
<td>7.9</td>
<td>15.8</td>
<td>Smith et al., 1986</td>
</tr>
<tr>
<td>Chamois (Rupicapra rupicapra)</td>
<td>4–11</td>
<td>55</td>
<td>3.4–5.3</td>
<td>18.2</td>
<td>Pemberton et al., 1989</td>
</tr>
<tr>
<td></td>
<td>7–55</td>
<td>41</td>
<td>4.6–6.5</td>
<td>9.7–17.0</td>
<td>Miller and Hartl, 1986</td>
</tr>
<tr>
<td>Roe deer (Capreolus capreolus)</td>
<td>12–62</td>
<td>41</td>
<td>3.5–7.9</td>
<td>14.6–19.5</td>
<td>Hartl and Reimoser, 1988</td>
</tr>
<tr>
<td>Arabian oryx (Oryx leucoryx)</td>
<td>24</td>
<td>19</td>
<td>2.4</td>
<td>10.5</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Moose (Alces alces)</td>
<td>85</td>
<td>18</td>
<td>5.2</td>
<td>16.7</td>
<td>This study</td>
</tr>
<tr>
<td></td>
<td>9–100</td>
<td>23</td>
<td>0.6–4.7</td>
<td>4.3–13.0</td>
<td>Ryman et al., 1980</td>
</tr>
<tr>
<td></td>
<td>165</td>
<td>19</td>
<td>1.7</td>
<td>15.8</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Mule deer (Odocoileus hemionus)</td>
<td>2</td>
<td>19</td>
<td>5.3</td>
<td>10.5</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Fallow deer (Dama dama)</td>
<td>18–118*</td>
<td>17</td>
<td>1.8</td>
<td>6.6</td>
<td>Hartl et al., 1986</td>
</tr>
<tr>
<td>Bison (Bos bison)</td>
<td>7</td>
<td>19</td>
<td>2.3</td>
<td>5.3</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Reindeer (Rangifer tarandus)</td>
<td>20</td>
<td>19</td>
<td>1.4</td>
<td>5.3</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Elk (Cervus canadensis)</td>
<td>25–200*</td>
<td>24</td>
<td>1.2</td>
<td>4.0</td>
<td>Cameron and Vyse, 1978</td>
</tr>
<tr>
<td></td>
<td>49</td>
<td>19</td>
<td>1.5</td>
<td>10.5</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Pronghorn (Antilocapra americanica)</td>
<td>5</td>
<td>19</td>
<td>1.1</td>
<td>5.3</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td>Red deer (Cervus elaphus)</td>
<td>3–80</td>
<td>34/35</td>
<td>0–3.0</td>
<td>0–13.8</td>
<td>Gyllensten et al., 1983</td>
</tr>
<tr>
<td>Ibex (Capra ibex)</td>
<td>27</td>
<td>19</td>
<td>4.7</td>
<td>15.8</td>
<td>Baccus et al., 1983</td>
</tr>
<tr>
<td></td>
<td>1–16</td>
<td>33</td>
<td>0–2.0</td>
<td>6.1</td>
<td>Stiwe and Scribner, 1989</td>
</tr>
<tr>
<td></td>
<td>46</td>
<td>38</td>
<td>2.3</td>
<td>5.3</td>
<td>Hartl, 1986</td>
</tr>
<tr>
<td></td>
<td>21</td>
<td>19</td>
<td>2.5</td>
<td>10.5</td>
<td>Granjon et al., 1990</td>
</tr>
<tr>
<td>Fallow deer (Dama dama)</td>
<td>88–368*</td>
<td>30</td>
<td>0</td>
<td>0</td>
<td>Pemberton and Smith, 1985</td>
</tr>
<tr>
<td>Caribou (Rangifer tarandus)</td>
<td>4</td>
<td>19</td>
<td>0</td>
<td>0</td>
<td>Baccus et al., 1983</td>
</tr>
</tbody>
</table>

*Extreme values (N ind., H, P) are given when different samples are distinguished.

*Range of individual numbers when they differ according to the loci sampled.

for instance, it should be possible to choose individuals that will restore variability at Ldh and Hb loci. However, this is advisable only if the material cost is not too high, since we do not know whether these characters are adaptively important [Hedrick et al., 1986]. 3) The allozyme survey would aid in determination of pedigrees [Ryder et al., 1981] where this information is lacking. The use of the 3 polymorphic loci found in our study makes this test more reliable, and has been used for the Mahazat As Said herd in a case where the sire’s identity was uncertain. Figure 3b illustrates this particular case, after the Mendelian transmission of the markers used had been verified on a number of cases of known parentages (example in Fig. 3a).
CONCLUSIONS

1. Eighty-five Arabian oryx analyzed at 18 loci were polymorphic for 17.6% of their protein-coding gene loci, and had a mean heterozygosity of 0.052.
2. Despite at least one bottleneck in its recent history, this species show levels of genetic variability typical of artiodactyls.
3. No significant differences between the two main samples (Saudi Arabia and Jordan) were found as far as genetic variability and differentiation are concerned.
4. Morphological as well as genetic (electrophoresis, DNA fingerprint) studies must continue to improve efficiency of captive-breeding programs by preserving as much variability as possible.
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