A STUDY OF GENETIC MARKERS IN THE SOUTH AFRICAN BLESBOK (*DAMALISCUS DORCAS PHILLIPSI*)

by

D. R. OSTERHOFF*, I. S. WARD-COX* and VALERIE EMLIE**

Abstract – During culling operations on blesbok from the Rietvlei Nature Reserve, 198 blood samples were obtained for study of genetic markers including haemoglobins, transferrins, amylases, albumins and carbonic anhydrases. All animals exhibited the same pattern.

Blood typing was performed by using goat reagents. These reagents could possibly be used to establish the relationship between the blesbok and the bontebok.

Immunoelectrophoresis was used to detect any immunological differences; 123 blesbok samples were tested against antibovine serum prepared in rabbits. The animals could be grouped into four distinguishable types according to the shape and presence of the alpha-2-macroglobulin and the "horizontal" band.

Introduction

The two antelope taxa *Damalesc dorcas phillipsi* (blesbok) and *Damalesc dorcas dorcas* (bontebok) have been the items of much taxonomical controversy in the past, in that there is no certainty as to whether they are two separate species or not. They do reveal certain morphological differences which may be attributed to their centres of distribution being far removed from each other, and it is a known fact that the bontebok approached extinction until it was saved (Van der Merwe, 1968). The blesbok, although not an animal found in great numbers, is in no danger of becoming extinct. The possibility has been expressed that the two groups were originally one while migrating southward, whereupon the bontebok settled in the Cape Province. This hypothesis has not been sufficiently substantiated, however, and in an attempt to do so an investi-

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gation of genetic markers and on the immunological patterns was initiated.

**Material and Methods**

During culling operations on blesbok from the Rietvlei Nature Reserve in the district of Pretoria, blood samples were obtained from 198 animals. These were subjected to serum protein analysis, in particular the transferrins, albumins, haemoglobins, amylases and carbonic anhydrases, using the well known technique of starch gel electrophoresis (Osterhoff, 1968).

The haemolytic test as described originally by Stormont and Cumley (1943) was employed in most of the procedures. Three main components in this test are (a) antigenic factors — blood factors, (b) antibodies present in different concentration in test sera or reagents and (c) complement, rabbit serum usually being the source.

In the test performed with the blesbok samples ten goat reagents were used i.e. sera containing antibodies produced by recipient goats after immunization with red cells from donor goats and absorbed with panels of goat cells to ascertain that monovalent reagents were actually obtained.

Furthermore, immuno-electrophoresis was performed on 123 of the blesbok serum samples. With the aid of immuno-electrophoresis, which is actually a further elaboration of precipitation tests in a gel matrix, the separation and identification of serum proteins can be achieved. The proteins are first electrophoresed in agar, and antiserum is then added to parallel troughs cut in the agar. The antibodies in the antiserum then form arcs of precipitation with the electrophoretically separated antigens, giving a twofold characterization of the antigens, by antigenic specificity and by electrophoretic mobility. Only anti-bovine serum was available and could successfully be used in the typing of blesbok sera.

**Results**

All 198 blood samples were analysed in the different electrophoretic systems used for haemoglobin, transferrin, amylase, albumin and carbonic anhydrase determination.

All samples exhibited the same pattern. Haemoglobin, amylase, albumin and carbonic anhydrase showing only one band, and transferrin also showing a one band pattern with a feint band migrating in front.

The red cells were then grouped using antisera against goat red cell antigens. Reactions were found with only three of the 10 antisera available, two giving strong results with the remaining one being weak but very specifically able to detect individual differences.

Similar conditions prevailed in the investigations done on the African buffalo by Osterhoff, Young and Ward-Cox (1970). The frequency distribution of blood factors appear in Table 1.
Table 1

The frequency of blood factors on blesbok red cells

<table>
<thead>
<tr>
<th>Blood factor</th>
<th>Frequency</th>
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<tbody>
<tr>
<td>G. 2</td>
<td>0.592</td>
</tr>
<tr>
<td>G. 6</td>
<td>0.833</td>
</tr>
<tr>
<td>G. 11</td>
<td>0.883</td>
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</tbody>
</table>

The following reaction phenomena were also noticed:

(i) 24 animals reacted with none of the antisera.
    9 animals reacted with 1 of the antisera.
    31 animals reacted with 2 of the antisera.
    56 animals reacted with 3 of the antisera.

(ii) In all cases where only one reaction was noted, it was with either G.6 or G.11, but never with G.2. Whenever both G.6 and G.11 were absent from the list of reactions, G.2 was also. It appears, however, that the presence of G.2 is more closely related to presence of G.11 than to that of G.6, as G.2 is always absent in the absence of G.11.

For the detection of any immunological differences, as evidenced by the immunoelectrophoretic patterns obtained, 123 blesbok samples were tested against an anti-bovine serum prepared in rabbits. Principal differences were found to lie in the region of the $\alpha_2$-macroglobulin arc and in the presence or absence of a horizontal arc probably a $\beta_2$M extending cathodically between the antigen well and the antibody trough. On this basis, variants could be subdivided into four main groups.

Figure 1 shows the immunoelectrophoretic variations found in the serum of blesbok.

Fig. 1. Immunoelectrophoretic variations found in the blood serum of blesbok.
The fact that no immunoelectrophoretic variations could be found in the albumin or transferrin bands supports the starch gel electrophoresis results for these components.

The percentage of animals falling into the four groups are shown in Table 2.

<table>
<thead>
<tr>
<th>Group</th>
<th>(a_2) macroglobulin band</th>
<th>Horizontal band</th>
<th>% of samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Continuous with the antigen well</td>
<td>Absent</td>
<td>11%</td>
</tr>
<tr>
<td>II</td>
<td>Not in contact with the antigen well</td>
<td>Absent</td>
<td>2%</td>
</tr>
<tr>
<td>III</td>
<td>As in I</td>
<td>Present</td>
<td>76%</td>
</tr>
<tr>
<td>IV</td>
<td>As in II</td>
<td>Present</td>
<td>11%</td>
</tr>
</tbody>
</table>

Discussion and Conclusions

It is an established fact that no two individuals of a species are completely alike. This individual variability can be established by the use of genetic markers, and varies a great deal from species to species.

Species are relatively stable complexes that persist over long periods of time, but they are not absolutely permanent. Modern geneticists have shown how species arise as a result of reproductive isolation. As Dobshansky and many others assume, geographical barriers play the chief role in this isolation. The study of phylogenetic relationships is basically the collection of evidence that leads to the general acceptance of theories of the common origin of species, in our case of the genus *Damaliscus*.

From the results obtained it can be concluded that, although the blesbok is homogeneous with respect to the investigated biochemical polymorphic factors of the blood, a definite mode of reaction of their red cells with antisera to goat red cell factors G.2, G.6 and G.11 exists. It is also evident from the results that three blesbok red cell factors are incurred in these reactions, namely one that cross-reacts with G.2 and G.6, one with G.2 and G.11 and one with G.2, G.6 and G.11. Making use of these facts, it would be possible to identify such a population at some later stage and even to establish the relationship between other supposed blesbok or bontebok herds in the same way that Osterhoff and Keep (1970) did with the black and white rhinoceros.
With regard to the immuno-electrophoretic studies it can be said that further investigations are needed to determine the possible mode of inheritance of the various types, e.g. whether codominant alleles are responsible. A similar investigation into the immuno-electrophoretic patterns of the bontebok may provide further insight into the problem and would also provide evidence to support or refute the contention that the bontebok are somehow related to the blesbok.

It will also be interesting to see whether an evolutionary trend is perhaps leading towards the loss of the horizontal band. The use of other antisera, a larger sample and a time study may clarify this point.

Acknowledgements

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REFERENCES


