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The Establishment of a Biomedical Reference Collection for a Captive Breeding and Restoration Programme

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Summary

The establishment of a collection of biomedical data and reference material by the National Avian Research Center, Abu Dhabi, for a bustard captive breeding and restoration programme is reported. The development and numerous practical uses of such a collection including investigations of disease are discussed. Comprehensive banks of biomedical data and material are essential for retrospective research projects allowing those managing captive breeding and restoration programmes to build up accurate biological profiles of free-living and captive populations in health and disease. The authors recommend that other zoological gardens or captive breeding and restoration programmes should consider the establishment of comparable biomedical collections.

Introduction

The National Avian Research Centre (NARC) was established by Royal Decree in September 1989, by H. H. Sheikh Khalifa bin Zayed Al Nahyan, Crown Prince of Abu Dhabi, United Arab Emirates (UAE). One of NARC's aims is to establish a captive breeding and restoration programme for the houbara bustard (*Chlamydotis undulata macqueenii*) and a professional team of scientists, aviculturalists and veterinarians has been recruited to achieve this aim. NARC also has successful breeding programmes for other species of African bustards including rufous-crested bustards (*Eupodotis ruficrista*), kori bustard (*Ardeotis kori*) and white-bellied bustards (*Eupodotis senegalensis*) and in the future may establish such programmes for black (*Eupodotis afra*) and Heuglin's bustards (*Neotis heuglini*).

The Veterinary Science Department of NARC is responsible for managing the health of bustards maintained at the different breeding and research facilities within the UAE, for conducting research aimed at improving the health of captive bustards and for investigating the diseases of wild houbara bustards. Unfortunately, the published literature on biomedical aspects of bustards is scarce (Bailey and others, in press). Monitoring the causes of morbidity and mortality of both free-living and captive bustards is essential to minimise the transfer of novel diseases between captive and free-ranging populations through re-introduction programmes (Hutchins et al 1991, Cooper, 1993, Kirkwood 1993, Munson and Cook 1993, Bailey and Samour, in press). Disease monitoring protocols of animals maintained in zoological collections should include provision for serum and tissue banking (Cooper, 1989; Cooper, 1993, Munson and Cook, 1993, Jacobson, 1993, Munson and Woodford, 1995; Bailey and Samour, in

press). Banks of biomedical tissues derived from both clinically normal and abnormal individuals are indispensable in retrospective investigations to determine the historical prevalence of diseases, exposure to infectious organisms within populations and also as a source of normal material for comparative studies.

A collection of biomedical material derived from houbara bustards and other species of captive bustards maintained by NARC was developed following the advice of one of the authors (Professor J. E. Cooper), a member the NARC International Advisory Committee (IAC). The latter body is composed of international specialists in the field of conservation and captive breeding and annually reviews all NARC's work programmes.

The main objective of this collection is to provide a central registry for research on health and diseases of bustards, but specimens are also available for traditional museum studies involving taxonomy, anatomy and genetics. Here we report the establishment and uses of a biomedical reference collection at the NARC and suggest that similar protocols could be successfully developed at other zoological gardens or captive breeding and restoration programmes.

Location and content of the reference collection

The collection is housed at the NARC Veterinary Science Department at the main Research Centre, Sweihan, Abu Dhabi and also at facilities in Al Ain Zoo. Material is derived from the species of bustards maintained in captivity by NARC and additionally from free-living houbara bustards captured in the UAE and Central Asia.

Data collection and storage

Customised clinical, surgical, laboratory and post-mortem examination forms were designed to record data. All data are entered into a computerised database, developed using Microsoft Access software (Microsoft Corporation, USA). The database was designed by NARC personnel and customised to ensure rapid inputting and retrieval of data. Data are readily available for individual birds and relate to medical history, breeding and husbandry practices.

Material collected from live birds

Clinical findings from live birds are recorded and entered into the database. Whole endoparasites and ectoparasites, red blood cells, serum, plasma and feathers are collected from living birds. Fig 1 illustrates the range of biomedical material and data that are archived from live birds.

Material collected from dead birds

The majority of material is collected at post-mortem examination and is summarised in figure 2. Morphometrics, organ biometrics and tissue samples are collected from dead birds in addition to the material and data described in figure 1 for live birds. A comprehensive description of the post-mortem examination technique for bustards is described elsewhere (Nicholls et al, in press). Preservation of tissues is achieved through the following;

neutral buffered formalin (tissues for histology investigations), ethanol (whole parasites), or freezing (tissues, serum, plasma, red cells or whole carcasses). Frozen material is stored in commercially available pathology freezer units (Denley, UK) maintained in standard chest freezers at - 20C. Whole bodies and remains of bodies are frozen as sources of skins and skeletal material. Data are recorded and entered into the database and Figure 3. is an example of a report illustrating the range of tissues stored

following a typical post-mortem examination. Other clinical, laboratory and post-mortem data are also accessible from this database.

Other material

Whole eggs and eggshell collected during the breeding season, colour transparency slides (of clinical conditions, surgical procedures, anatomical dissections, post-mortem findings and facilities), histology slides, blood films and parasitic identification slides are all maintained in the collection. Skeletal material has also been recovered from a set of buried carcasses. In the future it is intended to retain cultures of micro-organisms isolated from clinical, post-mortem and healthy bustards.

Uses of the Material Stored in the Collection

Retrospective analysis of biomedical records and material maintained in a Captive Breeding and Restoration Programme has a dual role; first to assess husbandry and biomedical management practices and secondly to target research projects at specific "problem areas" with the aim of allowing management to adapt and improve. Such research projects often need to access material that has been retained in a reference collection. For example, in 1995 it became apparent that fatty liver disease was a problem in bustards maintained in captivity in the UAE. A research project was undertaken to establish the incidence and severity of fatty liver disease by examining formalin-preserved livers and retrospective analyse of clinical and post-mortem records stored in the database. The project was able to establish risk factors for the disease and to suggest preventive and therapeutic measures based on the results.

The collection has already been used to supply material for a number of other projects and these illustrate the diverse range of uses of such a collection. The projects include;

1. Lead analysis of frozen tissue samples of a number of bustards suspected to have died of lead toxicity. A full description of lead toxicity in a flock of houbara bustards is described by Bailey et al (1995).
2. Submission of frozen serum samples for Newcastle disease haemagglutination inhibition analysis as part of a retrospective serological survey, to assess the effectiveness of the vaccination protocols (Wernery et al, in prep) and to assess historical exposure to paramyxovirus infections by the NARC bustard flock.
3. Use of material by a team of researchers from the National Museum of Scotland and the York Museum for the following purposes: material for environmental archaeologists, taxidermy specimens, taxonomical, anatomical, and physiological studies, and material for natural history exhibits.
4. Surveys of haemoparasites from stored blood smears (Howlett et al, in press).
5. Surveys of endoparasites in bustards collected post-mortem and during clinical examination (Jones et al, 1994).
6. Future research projects planned by NARC that will be accessing material and data stored in the collection include; genetic studies (frozen blood, frozen tissues or feather samples); virological surveys (frozen tissue/organ samples); toxicological/pesticide surveys (frozen fat/organ samples); histopathological studies (formalin samples); biochemistry analysis (frozen serum/plasma); Serological surveys against other avian infectious diseases (frozen serum samples); eggs from free-living and captive birds may be used for comparative studies on egg-shell thickness, porosity, mineral analysis and pesticide studies; studies of the micronutrients and trace elements of stored tissues.

Conclusions

The establishment of the reference collection and integration with a computerised database within one organisation permits the co-ordination of research work on this order of birds. There is an urgent need for more biomedical information on bustards, both to improve the health of birds maintained in captivity, and to minimise the transfer of diseases between wild and captive populations through re-introduction programmes. The importance of having access to biological samples that were collected at an earlier date in the investigation of wildlife diseases has been emphasised by a number of workers (Cooper, 1989; Cooper, 1993, Munson and Cook, 1993, Jacobson, 1993, Munson and Woodford, 1995; Remson, 1995; Bailey and Samour, in press). In the few instances where such collections have been established they have provided valuable material for investigations of species maintained in captivity. Examples of other collections include the Mascarene reference collection established at the Royal College of Surgeons of England (Cooper and Jones, 1986) and the Edward Elkan Collection of Lower Vertebrate Pathology (Williams, 1994). The development of the biomedical reference collection at the NARC illustrates the range of material that can be collected by a captive breeding programme and illustrates the many purposes for which it can be used. Most captive breeding programmes involve working with species which have been incompletely studied in the wild and have often never been studied in captivity. Biologists, veterinarians, and those involved with managing populations of captive and released animals are handicapped by the paucity of biomedical data on the species with which they work. Comprehensive banks of biomedical data and material are essential for retrospective research projects allowing those managing zoological collections to build up accurate biological profiles of their species in health and disease. Such collections may be of a wide or narrow range of tissues. Those working with other species should consider the establishment of comparable collections.

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Figure 2. Post-mortem examination protocol summary sheet.

Bird ID Give the bird a unique accession number e.g. AAZ/HB/170493/3.

History Record any observations or known previous history in the record sheet.

Radiography Take lateral and ventrodorsal radiographs of the whole bird prior to post-mortem. Take radiographs of any areas that warrant specific radiography e.g. fractured wing or suspected fractured neck.

Biometrics Weigh the bird and record biometric measurements prior to PM.

Blood samples If the bird is still alive awaiting euthanasia collect blood samples, EDTA samples for haematology, plasma for biochemistry and serum for serology. PM

organ measurements Record the weight and sizes of the following tissues; Weight - liver, abdominal fat, kidney, gizzard full, gizzard empty, spleen, pancreas

Length and width and activity - gonad

Length only - oesophagus, small intestine, large intestine, caeca, pancreas

Length and width - gizzard, spleen Archived tissues

Unless the bird is very decomposed, tissues must be retained in formalin or frozen for the reference collection. As a minimum the following tissues must be retained from any bird examined post mortem;

Formalin - brain, liver, kidney, spleen, lung, oesophagus, small intestine, large intestine, caeca, heart, pectoral muscle, sciatic nerve, air sac, gonad

Frozen - brain, liver, abdominal fat, kidney, lung, small intestine, large intestine, feathers and any carcass remains.

All abnormal tissues must be collected and stored in formalin