MALE REPRODUCTIVE FUNCTION AND THE DEVELOPMENT OF ASSISTED REPRODUCTION IN ENDANGERED FELIDS AND UNGULATES

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Among endengered species it is important to assess the impact of inbreeding on reproduction, to develop methods to bank genetic resources, particularly gametes and embryos, and to developed assisted reproductive techniques to facilitate genetic management. Inbreeding is known to cause deleterious effects upon reproduction and survival. The relation between inbreeding, heterozygosity, and reproductive fitness has been scarcely studied in endangered populations and there is considerable debate as to whether inbreeding (determined from pedigrees) or marker heterozygosity (calculated from microsatellites) better reflect inbreeding depression. In our experience, in captive breeding programmes of critically endangered Mohor gazelle and the Iberian lynx, marker heterozygosity (but not inbreeding coefficient) was associated with semen quality (proportion of normal sperm in the ejaculate). Thus, examination of heterozygosity-fitness correlations was found to be an effective way to detect inbreeding depression, particularly if the pedigree does not accurately reflect the history of inbreeding. We have also analyzed, among endangered gazelles, the relationship between inbreeding and sperm DNA integrity, and whether levels of sperm DNA fragmentation are associated with semen quality. There was an extremely high prevalence of sperm DNA damage in two gazelle species with high levels of inbreeding (*Gazella cuvieri* and *G. dama* mhorr) when compared to a species with low levels of inbreeding (*G. dorcas*), and to values previously reported for outbred populations. Increased DNA damage in sperm was associated with increased sperm head abnormalities and poor motility. The deleterious effects of inbreeding upon the paternal genome likely decrease male fertility and may cause genetic damage to future generations. Furthermore, sperm DNA damage may influence offspring survival; this possibility has not been explored before. We therefore examined maternal, paternal and individual factors that may influence offspring survival in the species of endangered gazelles with a very high inbreeding levels (*G. cuvieri*). We found that sperm DNA damage had an important impact upon offspring mortality with a significant interaction between this variable and maternal factors, so that offspring born to primiparous mothers were more likely to die if their father had high levels of sperm DNA damage. As part of our effort to develop semen evaluation protocols and assisted reproduction methods we have characterized, for both gazelles and the Iberian lynx, techniques for in vitro fertilization and artificial insemination. We have also developed methods of xenotransplantation of testicular tissue for the generation of spermatozoa from males that die before reaching reproductive maturity.