

## Evaluation of Porcine Zona Pellucida (PZP) Immunocontraception for Behavioral Effects in Feral Horses (*Equus caballus*)

David M. Powell

Department of Zoology, University of Maryland

Department of Zoological Research, National Zoological Park

### Introduction

One of the many challenges facing zoological parks and aquaria is the management and maintenance of genetically viable populations of endangered species. In theory these populations will eventually serve as reservoirs of genetic diversity for the reinforcement or re-establishment of wild populations of animals. To meet this goal, populations of animals must be managed efficiently and effectively given the constraints of space and resources. Breeding must be managed so as to maximize the production of genetically valuable individuals and minimize the numbers of either over-represented or genetically inferior individuals. In cooperation with the American Zoo and Aquarium Association, zoos and aquaria have developed a variety of tools to facilitate the development and execution of successful breeding programs. These tools include studbooks, population management plans, and species survival plans (SSPs) which all contribute, in one way or another, to making recommendations about allocation of captive space and pairing of individuals for reproduction. Coordination of breeding programs often involves many animals distributed over many institutions. In general the institutions participating in a breeding program agree on the means by which to achieve the program's objectives; however, unplanned pregnancies do occur and variation in litter or clutch sizes produces surplus individuals.

In most cases, the number of offspring required from an individual to maintain a genetically healthy captive population is significantly lower than the potential number of offspring it could produce. Thus, by their very design, programs like SSPs require animal managers to have a variety of tools available to allow for compliance to breeding recommendations (Asa *et al.*, 1996). These tools often must be applied to non-endangered species as well because they occupy valuable space or consume resources which may be needed for endangered species programs.

Two methods by which breeding can be managed are separation of the sexes and establishment of same sex groups of individuals. Separating the sexes may in some cases lead to negative effects on animal health and behavior (Conway, 1976). The practice of establishing same sex groups is in its infancy in zoological parks, though some groups have been established and maintained with success (Stoinski, pers. comm.). Given the current limitations of these methods, it appears that contraception is the best tool available for effective management of reproduction.

Research involving various contraceptive methods has traditionally been aimed at humans, companion animals, and wildlife. Much less is known about the efficacy and

following treatment and a trend toward increased affiliative behavior initiated by the females. All females had gained weight during the treatment period (range 17.3-47.6% increase over starting weights). They also found that perineal swellings never fully subsided, indicating that some tissue hypertrophy may have been permanent (Portugal & Asa, 1995). In a study of stumptail macaques (*Macaca arctoides*) Linn & Steklis (1990) found that during treatment with depo-medroxyprogesterone (Depo-Provera), another synthetic progestin, there was a decrease in rates of affiliation and a tendency for higher rates of aggression.

Hayes *et al.* (1996) observed the behavior of control and MGA implanted Rodrigues fruit bats during a pre-implantation period, one month post-implantation, and six months post-implantation. During the pre-implantation period there were no differences in activity budget between control and treated bats. One month post-implantation, they found that implanted bats moved more than control bats. At six months post-implantation, this trend had reversed; however both groups showed decreased movement compared to one month post-implantation. They also noted within group changes during these study periods (Hayes *et al.*, 1996). In an analysis of spatial relationships, they found that at one month post-implantation, treated bats spent significantly more time with a nearest neighbor than control bats. They found no effects of treatment on the age/sex structure of neighbors, nor in the number of displacements given or received. They also saw no effect of treatment on the rate of copulations. The authors suggested that the changes in activity budget between groups were most likely due to changes that occurred within groups and not to implants. Hayes *et al.* found that implanted females had gained weight (in excess of 8% of body weight), and seven of nine treated bats showed poor re-growth of hair and some scarring over the implant insertion site (1996).

McShea and colleagues (1997) evaluated the efficacy of porcine zona pellucida (PZP) immunocontraception (see below for details) and its impact on behavior of white-tailed deer (*Odocoileus virginianus*). Does were divided into a control group and two treatment groups. The two treatment groups differed in the total dose of PZP. Treatment with the smaller of two doses in this study did not prevent parturition, but it did significantly delay parturition dates. Parturition dates for contracepted females that did fawn were significantly later than the parturition dates of control does. As in the MGA studies discussed above, treated does showed significantly more weight gain than control does during the study period. McShea *et al.* (1997) found that males exhibited longer breeding seasons during the first year of their study, and in the second year they noted that the breeding season was not longer but had started over two weeks earlier. Among does, contracepted females were more active (exhibited more running/walking) than untreated does, though this difference was only evident during estrus and may have been due to the treated does undergoing one to two non-conceptive cycles before getting pregnant. Treated does also exhibited estrous periods later in the season than control does (McShea *et al.*, 1997).

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uncontracepted mares relative to the stallion in the harem band. One could imagine that contracepted mares might either a) be close to the stallion more often because they are continuously cycling during the breeding season and hence would show repeated periods of estrous behavior or b) be far from the stallion and on the periphery of the group because the stallion might find them unsuitable as mates. Finally I hypothesized that contracepted mares and uncontracepted mares would differ in social rank and the amount of aggression they gave and received.

## **Methods**

### **Study Animals**

With the assistance of Earthwatch volunteers, I conducted behavioral observations on a population of feral horses (*Equus caballus*). These horses live on Assateague Island National Seashore (A.S.I.S.), a 33 mile long barrier island running along the coasts of Maryland and Virginia. Observations were restricted to the Maryland herd which contains approximately 173 individuals and is essentially unmanaged except for population control.

Though equid social organization appears to be closely tied to ecology (Rubenstein, 1981), the typical social group in feral horses is the harem band which contains one breeding male, several reproductive mares, and their dependent offspring. Harem bands on Assateague occupy overlapping home ranges and are not characterized by territoriality. A dominance hierarchy exists within each harem band, and the stallion may not be the alpha animal (Haupt & Keiper, 1982). Among mares, rank correlates most highly with tenure (Seligsohn, 1987) but also very highly with age (Rutberg & Greenberg, 1990; Keiper & Sambraus, 1986). Female hierarchies tend to be stable with few reversals (Keiper & Sambraus, 1986 but see Berger, 1986). Nine bands of horses were observed June 1-September 5, 1997. These bands contained 43 sexually mature mares.

National Parks Service personnel and outside researchers had documented significant impacts by the horses on the island's vegetation. Concern over the size of the herd led to the development of a plan to reduce the population using the immunocontraceptive, porcine zona pellucida (PZP). Widespread inoculation of the herd with PZP began in 1994, though some mares had been treated previously as part of research on the reversibility of PZP. Of the 43 mares observed in this study, 24 were contracepted and 19 were uncontracepted during the breeding season of 1997, though all mares in the study had been contracepted for some period at some time in the past. Contracepted mares had been on contraception for an average of 2.54 consecutive breeding seasons (range 1-5). Eighteen of the 19 uncontracepted mares had been on contraception for the previous 3 breeding seasons (1994-1996) and were taken off treatment either to decrease potential side effects of longer term treatment or to allow for breeding.

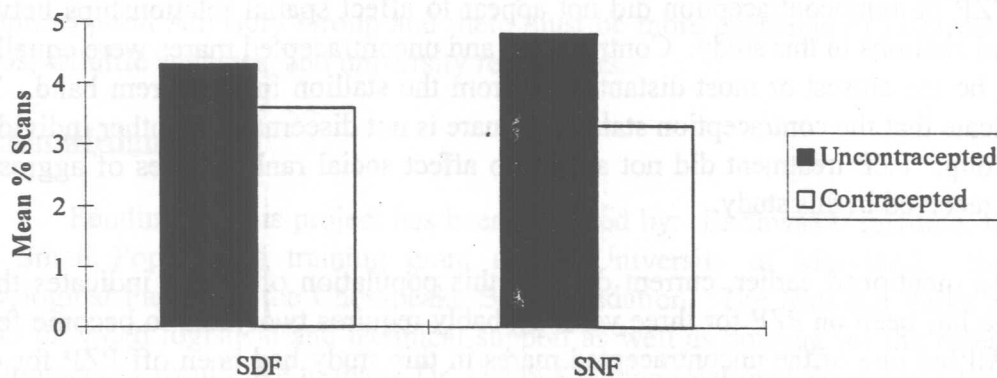
Table 1: Activity budgets of contracepted and uncontracepted mares.

| Behavior | % Time         |              | p*      |
|----------|----------------|--------------|---------|
|          | Uncontracepted | Contracepted |         |
| Graze    | 51.42          | 54.26        | p=.1012 |
| Rest     | 33.35          | 31.87        | p=.2764 |
| Stand    | 4.98           | 4.42         | p=.3654 |
| Locomote | 8.88           | 7.15         | p=.1388 |
| Groom    | 0.86           | 1.19         | p=.3517 |
| Social   | 0.47           | 1.10         | p=.2688 |

\*Mann-Whitney U Test

A mean percentage of scans in which a female was the nearest mare to the stallion and the most distant female from the stallion was calculated for each mare. These were then scaled by group size. There were no significant differences for either measure between contracepted and uncontracepted mares (stallion's nearest female:  $U=116$ ,  $p=.1661$ ; most distant female from stallion:  $U=144$ ,  $p=.6105$ , Figure 1).

Figure 1: Mean percentage of scans in which a mare was the stallion's nearest mare (SNF) and most distant mare (SDF), scaled by group size.



Mean dominance index was compared between contracepted and uncontracepted mares. Each of three mares which switched bands during the study were treated as two