Immunocontraception of mammalian wildlife: ecological and immunogenetic issues

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Abstract

Immunocontraception involves stimulating immune responses against gametes or reproductive hormones thus preventing conception. The method is being developed for the humane control of pest and overabundant populations of mammalian wildlife. This paper examines three fundamental issues associated with its use: (1) the difficulties of obtaining responses to self-antigens, (2) the likely evolution of genetically based non-response to immunocontraceptive agents, and (3) the possible changes in the array of pathogens possessed by the target species after generations of immunocontraception. Our review of the literature demonstrates that the barriers to an effective immunocontraceptive are at present very basic. Should they be overcome, the effects of immunocontraception on the immunogenetic constitution of wildlife populations through the selection for non-responders must be examined. We suggest that the attempt to use the animal's own immune system to modulate reproduction may be incompatible with the basic biological function of protection against infectious disease. Research programs on mammalian immunocontraception should involve measurement of the heritability of non-response and an assessment of the likely change in the response of the contracepted population to possible pathogens.

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Introduction

The regulation of human and animal population numbers constitutes a difficult and largely unsolved contemporary problem. In the developed world, steroid contraceptives for humans are both widely used and efficacious. Elsewhere they are too costly. The development of less expensive methods is considered necessary (Aitken *et al.* 1993). One such method is immunocontraception, i.e. the vaccination against sperm, eggs, or reproductive hormones to prevent either fertilization or the production of gametes. Attempts to design human immunocontraception have a long history (Joshi 1973, Stevens 1975, Basten 1988, Gupta & Talwar 1994). The targets include sperm antigens, oocyte antigens, especially zona pellucida proteins (PZP), gonadotropin riboflavin carrier protein, gonadotropins and gonadotropin releasing hormones (Delves *et al.* 2002). The most advanced method involves immunization against human chorionic gonadotropin, in reality a method of very early pregnancy termination (Baird 2000). It now seems likely that problems associated with autoimmune disease and variability of response will prevent any widespread use of immunocontraception in humans in the foreseeable

future (Rao 2001, Aitken 2002). Women's health advocates have objected to all forms of immunocontraception because of perceived health risks and the potential for political abuse of the vaccine (Richter 1994). Human male immunocontraceptives have received much less attention and do not appear to be feasible in the near future (Delves *et al.* 2002).

Immunocontraceptives for wild animals have a different objective than those for humans. Their main aim is to check population growth rather than to contracept particular individuals. If some animals are irreversibly sterilized so much the better whereas such an effect in human medicine would be ethically most undesirable. Immunocontraceptives for animals are ostensibly humane and could potentially be used on the large scale required for wildlife population regulation. Research progress to date has been reviewed in Tyndale-Biscoe (1991, 1994), Barber & Fayrer-Hosken (2000), Barlow (2000), and Cooper & Herbert (2001). Three fundamental questions remain to be addressed: (1) Can sufficiently strong immune responses be provoked against the antigens (immunogens) of gametes or reproductive hormones to cause contraception in a proportion of animals large enough for effective

population management? (2) How rapidly will variation in these responses lead to the evolution of failure to respond to the immunocontraceptive agent? (3) What will be the ecological consequences of the likely changes to the immunogenetic constitution of the population as a result of selection for non-responders? In particular, will the endemic pathogens of the species change? There is considerable information which allows us to answer at least in part the first two questions. The third is of fundamental importance but even a preliminary answer is not possible at present.

Target species

Population control of native and exotic mammals is generally justified by environmental degradation, competition with and predation on native wildlife, conflicts with humans over food production, potential spread of pathogenic infectious diseases and the possibility of population crashes of over-abundant fauna or of wildlife populations near urban areas. Although still in its infancy, immunocontraception is regarded as being more humane than the traditional methods of wildlife population control, such as shooting, trapping, poisoning, or pathogenic agents and its use has strong support from influential animal welfare agencies worldwide (Oogjes 1997, Grandy & Rutberg 2002). Table 1 lists mammalian species for which immunocontraception is being investigated and for which the method could be applied. In all these species, there are at present no completely efficacious, cost-effective, or socially acceptable methods for population regulation available.

Immune responses to self-antigens

Responses to self-antigens are unusual and mainly weak. This constitutes a major barrier to the development of an immunocontraceptive. Table 2 summarizes data on attempts to induce immunocontraception using a variety of antigens in 14 mammalian species. The data in Table 2 show that in most cases a significant proportion of the population is not contracepted by administration of the immunocontraceptive antigen. The reason for this could be either genetic or environmental. In either case it indicates that a fraction of the population will continue to breed despite the administration of the contraceptive. In most cases, there is likely to be at least in part genetic causes underlying lack of response. If so, the genes for lack of response will be selected for and in a comparatively small number of generations most of the population will be non-responsive. This implies that the immunocontraceptive can be useful for only a short period of time.

All studies summarized in Table 2 involve the use of some kind of adjuvant, i.e. a substance or array of substances designed to enhance the immune response. There are no reports of successful immunocontraception without some form of adjuvant. Moreover, most

immunizations were boosted at least once (see Table 2). The need for multiple injections and the dependence upon adjuvant to achieve the necessary level of response renders the whole approach impractical at present. The most commonly used adjuvant, Freund's Adjuvant, also induces a range of undesirable side effects and its use is being challenged on animal welfare grounds (Leenaars *et al.* 1994, 1998). There is at present no feasible or acceptable method of promoting responses to self-antigens sufficient to cause immunocontraception.

Jackson *et al.* (2001) attempted to overcome the problem of lack of immune response to self-antigens in the absence of adjuvant by inserting the cytokine interleukin-4 into mousepox virus with the intention of increasing the humoral response. The virus was then inserted into the mice with the unwelcome outcome that the mice all died very quickly. This work caused alarm because of the possibility that this technology could lead to a method for simple conversion of relatively innocuous viruses into lethal ones, which could be used in biological warfare (Finkel 2001).

Another possible problem with virus-vectored immunocontraception is the potential for the horizontal transfer of the immunocontraceptive gene into viruses affecting other species (Becker 2000). While it may be possible to create genetically modified organisms without adverse effects on the target animals, the effects they might have on related species they come in contact with make any use of this approach questionable.

Variation in response and genetic change

Variation in response to biocontrol agents is a widespread phenomenon. This variation has frequently led to evolution of a degree of resistance so that the agent is no longer useful. The evolution of resistance to insecticides has been reviewed by McKenzie (1996). He draws a distinction between biocontrol agents with responses within the phenotypic range and those with responses outside the phenotypic range of the target species. An agent which initially kills all members of the target species is acting outside the normal phenotypical range, while one which kills only a fraction of the population is acting within the normal phenotypical range. He points out that when resistance appears in the former case it is frequently monogenic, while in the latter case a number of different genetic regions are involved, i.e. it is probably multifactorial. The basic genetic parameter to be estimated in either case is heritability, i.e. the extent to which genetic variation is controlled by genetic as opposed to environmental factors. The relative fertility of the immunocontracepted animals in Table 2 is > 10% in 27 out of the 32 data sets. A proportion of non-responders is characteristic of most species. Only three species (Tammar wallaby, Fallow deer and Norway rat) out of 14 had no non-responders (Table 2). Following McKenzie's (1996)

Table 1 Immunocontraception: target species and justification for fertility control.

Species	Location	Management issues	Population control needs	References
K-selected species				
Koala	Australia; local overpopulations	Habitat degradation; likely destruction of own habitat; highly regarded species	Control methods with public acceptance	Martin & Handasyde (1999)
African elephant	Southern Africa	Habitat degradation; public safety and health concerns; highly regarded species	Control methods with public acceptance	Hanks (2001)
Wild horse	USA, Australia	Habitat degradation; conflicts with livestock, timber and mining industry interests (USA); highly regarded species	Control methods with public acceptance	Berger (1986), Dobbie <i>et al.</i> (1993), Furbish & Albano (1994)
White-tailed deer	USA	Habitat degradation; public safety and health concerns; high frequencies of deer-vehicle collisions; crop and garden damage	Control methods with public acceptance	McShea <i>et al.</i> (1997), Warren (1997)
Feral donkey/Burro	USA, Australia, Africa	Habitat degradation; public safety and health concerns; highly regarded species	Broad-scale control over large, remote and inaccessible areas (Australia)	McCool (1981), Berger (1986), Freeland & Choquenot (1990)
Brush-tail possum	New Zealand; major introduced pest species	Habitat degradation (New Zealand); public, livestock and wildlife health concerns; reservoir for bovine tuberculosis	Broad-scale control over large, often inaccessible, areas (New Zealand). Alternative to poison baits (1080)	Montague (2000)
Macropods	Australia	Public, livestock and wildlife health concerns; high frequencies of kangaroo-vehicle collisions; highly regarded species	Control methods with public acceptance	Dawson (1995), Pople & Grigg (1999)
European red fox	Australia; major introduced pest species	Predation on native wildlife (Australia); public, livestock and wildlife health concerns	Control over continental area (Australia). Alternative to poison baits (1080)	Saunders <i>et al.</i> (1995)
Pinnipeds	Worldwide	Possible contribution to the depletion of fish stocks	Contraception suggested as humane alternative to culling	Butterworth <i>et al.</i> (1988), Brown <i>et al.</i> (1996), Mohn & Bowen (1996)
Feral cat	Worldwide; major introduced pest species in Australia	Predation on native wildlife (Australia); public and wildlife health concerns	Control over continental area (Australia) Alternative to poison baits (1080)	Newsome (1991), Bomford <i>et al.</i> (1996), Mahlow & Slater (1996)
Feral dog	Worldwide	Public and livestock safety and health concerns; predation on native wildlife (Australia)	Control methods with public acceptance	Fleming <i>et al.</i> (2001), Sabeta <i>et al.</i> (2003)
Feral pig	Worldwide; major introduced pest species in Australia	Habitat degradation; damage to economic resources; public, livestock and wildlife health concerns	Control methods with public acceptance	Choquenot <i>et al.</i> (1996)
Badger	UK	Public, livestock and wildlife health concerns; reservoir for bovine tuberculosis	Control methods with public acceptance	Krebs <i>et al.</i> (1998), Donnelly <i>et al.</i> (2003)
Grey squirrel	UK; introduced species	Habitat degradation; threat to the native Red squirrel	Control methods with public acceptance	Moore <i>et al.</i> (1997)
r-selected species				
European rabbit	Worldwide; major vertebrate pest species in Australia	Habitat degradation; major cost to agriculture; public, livestock and wildlife health concerns	Broad-scale control over large areas	Lawson (1995), Williams <i>et al.</i> (1995)
Rodents	Worldwide	Major damage to economic resources, incl. crops, pastures, stored grain, livestock, buildings and infrastructure; public, livestock and wildlife health concerns	Broad-scale control over large areas. Species-specific alternatives to rodenticides	Caughley <i>et al.</i> (1998), Chambers <i>et al.</i> (1999), Seamark (2001)

argument, this implies that non-response is likely to be multi-factorial in genetic terms. There are no data which will allow estimates of the heritability of non-response to immunocontraception in any of the species in Table 2. We are unable to predict the rate at which this characteristic

will increase in any one of these populations under the selective influence of immunocontraception. However, some idea of the likely change per generation given the initial frequency of non-responders can be found using Falconer's (1965) model for threshold characters (Table 3).

Table 2 Relative fertility of immunocontracepted females in 14 mammalian species.

Species	Immunogen	Adjuvant	No. of immunizations	Reproductive performance (control, treated)	Statistical significance ^a	Reduction in relative fertility (%) ^b	Reference
Baboon	LDH-C ₄ + promiscuous epitope	CGP11637 emulsified w. Squalene:Arlacel -A (4:1)	3	Offspring/females 10/13, 4/14	$P < 0.02$	62	O'Hern <i>et al.</i> (1997)
Brushtail possum	Whole sperm	Freund's complete (FCA), Freund's incomplete (FIA) in boosters	3	Offspring/females 12/16, 2/16	$P < 0.001$	83	Duckworth <i>et al.</i> (1998)
Tammar wallaby	Porcine ZP	FCA, FIA in boosters	5	Offspring/females 4/6, 0/6	$P < 0.05$ ($n=6$)	100	Kitchener <i>et al.</i> (2002)
African elephant	Porcine ZP	Adjuvant used, type not given	3	Offspring/females 16/18, 8/18	$P = 0.005$	50	Fayrer-Hosken <i>et al.</i> (2000)
First schedule	Porcine ZP	Adjuvant not mentioned	2	Offspring/females 2/10 (no true control)	$P = 0.001$	77	Fayrer-Hosken <i>et al.</i> (2000)
Second schedule	Porcine ZP	FCA, FIA in boosters	3–4	Pregnancy rate 3/6, 1/14	Not stated	85	Kirkpatrick <i>et al.</i> (1991)
Wild horse	Porcine ZP	FCA, FIA in boosters	3–4	Pregnancy rate 3/6, 1/14	Not stated	85	Kirkpatrick <i>et al.</i> (1991)
White-tailed deer	Porcine ZP	FCA, FIA in booster	2	Fawns/doe years 30/16, 4/16	$P < 0.0001$	87	Miller <i>et al.</i> (2000a)
First schedule	Porcine ZP	FCA, FIA in booster	2	Fawns/doe years 30/16, 19/14	$P < 0.05$	28	Miller <i>et al.</i> (2000a)
Second schedule	RC55	FCA, FIA in booster	2	Fawns/doe years 30/16, 11/8	$P < 0.01$	27	Miller <i>et al.</i> (2000a)
Third schedule	RC75a	FCA, FIA in booster	2	Fawns/doe years 30/16, 16/8	$P > 0.05$	0	Miller <i>et al.</i> (2000a)
Fourth schedule	Combined antigens	FCA, FIA in booster	2	Fawns/doe years 35/19, 5/24	$P < 0.01$	89	Miller <i>et al.</i> (2000b)
White-tailed deer	KLH-GnRH	FCA, FIA in boosters	2–4	Fawns/doe years 35/19, 25/57	$P < 0.01$	76	Miller <i>et al.</i> (2000c)
White-tailed deer	Porcine ZP	FCA, FIA in boosters	2–3	Fawns/doe years 35/19, 25/57	$P < 0.01$	76	Miller <i>et al.</i> (2000c)
White-tailed deer	GnRH	FCA, FIA in boosters	3	Fawns/doe years 110/90, 36/118	$P < 0.0005$	75	Curtis <i>et al.</i> (2002)
First schedule	GnRH	FCA, FIA in boosters	3	Fawns/doe years 72/56, 10/60	$P < 0.0005$	87	Curtis <i>et al.</i> (2002)
Second schedule	Porcine ZP	FCA, FIA in boosters	3	Pregnancy rate 322/334, 0/22	$P < 0.0001$	100	Fraker <i>et al.</i> (2002)
Fallow deer	SpayVac	FCA	1	Pregnancy rate 322/334, 0/22	$P < 0.0001$	100	Fraker <i>et al.</i> (2002)
Burro	Porcine ZP	FCA, FIA in boosters	2–3	Offspring/females 6/11, 1/16	$P < 0.05$	88	Turner <i>et al.</i> (1996)
Grey seal	SIZP (SpayVac)	FCA	1	Pups/female 2.76, 0.22	$P < 0.001$	92	Brown <i>et al.</i> (1996, 1997)
Tule elk	Porcine ZP	FCA, FIA in boosters	3–4	Calves/cow years 53/91, 5/104	Not stated	91	Shideler <i>et al.</i> (2002)
Cat	Porcine ZP	FCA, FIA in boosters	5	Pregnancy rate 2/2, 1/5	Not stated	50	Ivanova <i>et al.</i> (1995)
Cat	SpayVac	FCA	1	Mean litter size 5.2, 4.5	$P = 0.8859$	13	Gorman <i>et al.</i> (2002)
First schedule	SpayVac	Alum	1	Mean litter size 5.2, 4.4	$P = 0.8859$	15	Gorman <i>et al.</i> (2002)
Second schedule	SpayVac	FCA, FIA in boosters	3	Mean litter size 7.4, 7.0	Not stated	5	Kerr <i>et al.</i> (1999)
European rabbit	Myxoma vectored ZPB	FCA, FIA in boosters	3	Mean litter size 7.4, 7.0	Not stated	5	Kerr <i>et al.</i> (1999)
Norway rat	MZPP/KLH	FCA, FIA in boosters	3	Pregnancy rate 7/8, 4/8	$P > 0.05$	40	Miller <i>et al.</i> (1997)
First schedule	MZPP/KLH	FCA, FIA in boosters	3	Pregnancy rate 7/8, 0/8	$P < 0.004$	100	Miller <i>et al.</i> (1997)
Second schedule	GnRH/KLH	FCA, FIA in boosters	5	Pregnancy rate 8/15, 7/30	$P = 0.046$	56	Hardy <i>et al.</i> (2002b)
Wild mouse	KLH-mZP3	FCA, FIA in boosters	5	Pregnancy rate 8/15, 7/30	$P = 0.046$	56	Hardy <i>et al.</i> (2002b)
BALB/c mouse	Murine rFA-1	FCA, FIA in boosters	4	Mean litter size 8.9, 3.2	$P < 0.0001$	64	Naz & Zhu (1998)
First schedule	Murine rFA-1	FCA, FIA in boosters	4	Mean litter size 8.6, 9	$P > 0.05$	0	Naz & Zhu (1998)
Second schedule	Murine rFA-1	FCA, FIA in boosters	4	Mean litter size 8.6, 9	$P > 0.05$	0	Naz & Zhu (1998)
BALB/c mouse	sp56FLAG	FCA, FIA in boosters	6	Offspring/females 55/14, 12/5	$P = 0.017$	39	Hardy & Mobbs (1999)
First schedule	sp56FLAG	FCA, FIA in boosters	6	Offspring/females 55/14, 12/5	$P = 0.017$	39	Hardy & Mobbs (1999)

Table 2 Continued.

Species	Immunogen	Adjuvant	No. of immunizations	Reproductive performance (control, treated)	Statistical significance ^a	Reduction in relative fertility (%) ^b	Reference
Second schedule	sp56FLAG	FCA, FIA in boosters	4	Offspring/females 55/14, 19/5	Not stated	3	Hardy & Mobbs (1999)
BALB/c mouse							
First schedule	MBP-polyepitope A	FCA, FIA in boosters	4	Mean litter size 5.2, 3.3	Not stated	37	Hardy <i>et al.</i> (2002a)
Second schedule	MBP-polyepitope B	FCA, FIA in boosters	4	Mean litter size 5.2, 2.1	$P < 0.05$	60	Hardy <i>et al.</i> (2002a)
Third schedule	6XHis-polyepitope A	FCA, FIA in boosters	4	Mean litter size 6.5, 6.3	Not stated	3	Hardy <i>et al.</i> (2002a)

Investigations have been conducted on approximately 70 species. This table includes only true experiments, i.e. studies with an immunized group compared with a control group.

^aThe P values are those given in the references cited. ^bRelative fertility is defined as the mean no. of offspring for females in the vaccinated group divided by the same figure for the control group (i.e. unimmunized females).

Reproduction is a good example of a threshold character; it is an all-or-none attribute which can be affected by a variety of underlying genetic and environmental factors. If heritabilities are high, rapid selection occurs. This is shown by the high percentage of non-responders that occur within one generation (Table 3).

A limited place for immunocontraception in wildlife management could be in species with long generation times. Genetic changes in them, if they occur, will take decades. Claims have been made for the potential efficacy of immunocontraception for African elephants (Fayrer-Hosken *et al.* 2000, Delsink *et al.* 2002), although this view has been challenged on demographic grounds (Pimm & van Aarde 2001). Long-term studies on immunocontraception in wild horses report 78–94.2% contraceptive efficacy (Kirkpatrick *et al.* 1995, Turner *et al.* 1997, 2002, Turner & Kirkpatrick 2002).

Zoo animals are convenient for immunocontraception studies of wild species, because of their long-term accessibility, although the small numbers usually available make controls hard to find. This is illustrated by an investigation involving 27 females from 10 felid species. Immunization with PZP and Freund's Complete Adjuvant gave several kinds of adverse reaction but no convincing evidence of an effect upon fertility (Harrenstien *et al.* 2004).

Table 3 Predicted proportion of non-responder daughters after one generation of selection by immunocontraception of mothers given various heritabilities (after Falconer 1965).

Non-responder mothers (%)	Heritability (%)			
	100	80	60	50
5	15	11	10	8
10	23	19	16	15

The prediction has been arrived at by entering the table in Falconer (1965) which relates heritability for a threshold trait to incidences in parent and offspring. The response to selection (predicted percentage of non-responders) obtained has been halved because selection is being carried out on one sex.

Delves & Roitt (2005) review attempts to immunocontracept mammals and conclude that GnRH is the most promising target, because of its evolutionary conserved sequence.

Immunogenetic issues

Infectious diseases are assumed to be one of the main classes of selective forces which act upon genes controlling immune responses (Klein *et al.* 1993). Immunocontraceptive agents also have the potential to influence the genetic constitution of a population with respect to the ability to mount immune responses. The two are similar in that pathogens which cause plagues and immunocontraceptive agents are both capable of exerting very strong selective pressure with the potential for rapid genetic change. However, they differ in two important respects. First, most pathogens are cellular and antigenically more complex than most immunocontraceptive agents, which consist of one or a few proteins and in some cases associated carbohydrate. Second, they have opposite directions of selection; resistance to a pathogen involves a positive response, whereas resistance to an immunocontraceptive involves non-response. The consequences of this kind of selection imposed by an immunocontraceptive agent require study. It seems likely that it will alter the immunogenetic constitution of the target species. The existence of genes governing response to pathogens is well documented in humans for malaria (Hill 2001), tuberculosis (Blackwell 1998, Bellamy 2003), and HIV (McNicholl & Cuenco 1999, Carrington & O'Brien 2003). Relevant examples are found in New Zealand Red Deer in which susceptibility to tuberculosis has high heritability (Mackintosh *et al.* 2000) and in the NRAMP1 association with the human response to leishmaniasis (Bucheton *et al.* 2003). The complexities of the co-evolution of pathogens and hosts and its biomedical significance are beginning to be unraveled (Woolhouse *et al.* 2002). Both experimental (Lively & Dybdahl 2000) and theoretical analyses (e.g. Nowak

& May 1994) point to the inherent difficulties of prediction of the course of these interactions. Prediction and detection of the ecopathological consequences of the use of immunocontraception of wild animals will also be made difficult by the spread of emerging infectious diseases as a result of human activity (Daszak *et al.* 2000).

The effect of immunocontraception upon genetic diversity could be significant. There is the possibility that restriction of breeding to a small group of animals which are closely related will result in localized inbreeding. This will be especially likely if their capacity to resist the immunocontraceptive is the result of shared uncommon genotype. Acevedo-Whitehouse *et al.* (2003) have shown that in California sea lions, inbreeding is associated with a wide range of diseases. They suggest that inbred individuals could act disproportionately as reservoirs of infectious agents.

Selection based upon immune responses could be on one of two parts of the genome: the MHC (major histocompatibility complex) region which governs responses to specific immunogens, or other genes, e.g. *NRAMP* which govern the functioning of the immune system in general (Bellamy 1999). The tightly linked *MHC* genes and the resultant linkage disequilibrium mean that selection on one gene will result in changes in gene frequencies at other loci. This could either raise or lower susceptibility to other pathogens. Understanding of the non-MHC genetic component of variability of the immune response is much less advanced than for the MHC component. This understanding is needed to attempt any predictions concerning immunocontraception-based selection.

Experimental approaches to this question have until now been very difficult. The existence of genetic maps of some wild animals (e.g. Tammar wallaby (Zenger *et al.* 2002)) may now allow a genomic approach, in which whole genome DNA typing may allow the identification of changes in gene frequency which accompany the application of immunocontraception or a pathogen, through comparing treated and control populations. A concomitant survey of pathogens in the two groups may identify susceptibility regions, whose existence could be further tested in lab-based investigation. The genome would thus be assayed for these genes, and test the extent to which the same genes are involved in responses to different pathogens and to immunocontraception.

A good model system to address these questions is wildlife tuberculosis which is of economic significance in several countries, e.g. Britain (Delahay *et al.* 2002), New Zealand (Buddle *et al.* 2002), and the United States (Palmer *et al.* 2002). Considerable information on the genetic control of response to mycobacterial antigens is available (North & Medina 1998, Kramnik *et al.* 2000, Bellamy 2003). The possibility of obtaining results relevant to human mycobacterial susceptibility may also encourage use of this system.

We conclude that attempting to use the immunological system to modulate reproduction could be

incompatible with the basic biological function of resisting pathogens. We have not discussed some of the practical issues. For example, all fertility control methods have the problem of delivery of the control agent. Highly valued animals must be treated without harming them. When this is the case, fertility control methods with fewer concomitant problems, such as surgical sterilization or the use of steroids or gonadotropin-based hormones, would be competitive with immunocontraception (Cooper & Herbert 2001).

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References

- Acevedo-Whitehouse K, Gulland F, Denise G & Amos W 2003 Inbreeding: disease susceptibility in California sea lions. *Nature* **422** 35.
- Aitken RJ 2002 Immunocontraceptive vaccines for human use. *Journal of Reproductive Immunology* **57** 273–287.
- Aitken RJ, Paterson M & Koothan PT 1993 Contraceptive vaccines. *British Medical Bulletin* **49** 88–99.
- Baird DT 2000 Overview of advances in contraception. *British Medical Bulletin* **56** 704–716.
- Barber MR & Fayrer-Hosken RA 2000 Possible mechanisms of mammalian immunocontraception. *Journal of Reproductive Immunology* **46** 103–124.
- Barlow ND 2000 The ecological challenge of immunocontraception: editor's introduction. *Journal of Applied Ecology* **37** 897–902.
- Basten A 1988 Birth control vaccines. *Baillière's Clinical Immunology and Allergy* **2** 759–774.
- Becker Y 2000 Evolution of viruses by acquisition of cellular RNA or DNA nucleotide sequences and genes: an introduction. *Virus Genes* **21** 7–12.
- Bellamy R 1999 The natural resistance-associated macrophage protein and susceptibility to intracellular pathogens. *Microbes and Infection* **1** 23–27.
- Bellamy R 2003 Susceptibility to mycobacterial infections: the importance of host genetics. *Genes and Immunity* **4** 4–11.
- Berger J 1986 *Wild Horses of the Great Basin: Social Competition and Population Size*, Chicago: University of Chicago Press.
- Blackwell JM 1998 Genetics of host resistance and susceptibility to intramacrophage pathogens: a study of multicausal families of tuberculosis, leprosy and leishmaniasis in north-eastern Brazil. *International Journal for Parasitology* **28** 21–28.
- Bomford M, Newsome A & O'Brien P 1996 Solutions to feral animal problems: ecological and economic principles. In *Conserving Biodiversity: Threats and Solutions*, pp 202–209. Eds RA Bradstock, DA Keith, RT Kingsford, D Lunney & DP Sivertsen. Sydney: Surrey Beatty and Sons.
- Brown RG, Kimmins WC, Mezei M, Parsons JL, Phojjajak B & Bowen WD 1996 Birth control for grey seals. *Nature* **379** 30–31.

- Brown RG, Bowen WD, Eddington JD, Kimmins WC, Mezei M, Parsons JL & Pohajdak B 1997 Evidence for a long-lasting single administration contraceptive vaccine in wild grey seals. *Journal of Reproductive Immunology* **35** 43–51.
- Bucheton B, Abel L, Kheir MM, Mirgani A, El-Safi SH, Chevillard C & Dessein A 2003 Genetic control of visceral leishmaniasis in a Sudanese population: candidate gene testing indicates a linkage to the NRAMP1 region. *Genes and Immunity* **4** 104–109.
- Buddle BM, Skinner MA, Wedlock DN, Collins DM & de Lisle GW 2002 New generation vaccines and delivery systems for control of bovine tuberculosis in cattle and wildlife. *Veterinary Immunology and Immunopathology* **87** 177–185.
- Butterworth DS, Duffy DC, Best PB & Bergh MO 1988 On the scientific basis for reducing the South African seal population. *South African Journal of Science* **84** 179–188.
- Carrington M & O'Brien SJ 2003 The influence of HLA genotype on AIDS. *Annual Review of Medicine* **54** 535–551.
- Caughley J, Bomford M, Parker B, Sinclair R, Griffiths J & Kelly D 1998 *Managing Vertebrate Pests: Rodents*, Canberra: Bureau for Rural Sciences.
- Chambers LK, Lawson MA & Hinds LA 1999 *Biological control of rodents – the case for fertility control using immunocontraception*, *Ecologically-Based Management of Rodent Pests*, Canberra: Australian Centre for International Agricultural Research.
- Choquenot D, McIlroy J & Korn T 1996 *Managing Vertebrate Pests: Feral Pigs*, Canberra: Bureau of Rural Sciences.
- Cooper DW & Herbert CA 2001 Genetics, biotechnology and population management of over-abundant mammalian wildlife in Australasia. *Reproduction, Fertility, and Development* **13** 451–458.
- Curtis PD, Pooler RL, Richmond ME, Miller LA, Mattfield GF & Quimby FW 2002 Comparative effects of GnRH and porcine zona pellucida (PZP) immunocontraceptive vaccines for controlling reproduction in white-tailed deer (*Odocoileus virginianus*). *Reproduction (Cambridge, England) Supplement* **60** 131–141.
- Daszak P, Cunningham AA & Hyatt AC 2000 Emerging infectious diseases of wildlife—threats to biodiversity and human health. *Science* **287** 443–449.
- Dawson TJ 1995 *Kangaroo: Biology of the Largest Marsupial*, Sydney: University of New South Wales Press.
- Delahay RJ, De Leeuw AN, Barlow AM, Clifton-Hadley RS & Cheeseman CL 2002 The status of *Mycobacterium bovis* infection in UK wild mammals: a review. *Veterinary Journal* **164** 90–105.
- Delsink AK, van Altena JJ, Kirkpatrick J, Grobler D & Fayrer-Hosken RA 2002 Field applications of immunocontraception in African elephants (*Loxodonta africana*). *Journal of Reproduction and Fertility. Supplement* **60** 117–124.
- Delves PJ & Roitt IM 2005 Vaccines for the control of reproduction-status in mammals and aspects of comparative interest. *Developments in Biologicals* **121** 265–273.
- Delves PJ, Lund T & Roitt IM 2002 Antifertility vaccines. *Trends in Immunology* **23** 213–219.
- Dobbie WR, Berman DMK & Braysher ML 1993 *Managing Vertebrate Pests: Feral Horses*, Canberra: Bureau of Resource Sciences.
- Donnelly CA, Woodroffe R, Cox DR, Bourne J, Gettinby G, Le Fevre AM, McNerney JP & Morrison WI 2003 Impact of localized badger culling on tuberculosis incidence in British cattle. *Nature* **426** 834–837.
- Duckworth JA, Buddle BM & Scobie S 1998 Fertility of brushtail possums (*Trichosurus vulpecula*) immunised against sperm. *Journal of Reproductive Immunology* **37** 125–138.
- Falconer DS 1965 The inheritance of liability to certain diseases, estimated from the incidence among relatives. *Annals of Human Genetics* **29** 51–76.
- Fayrer-Hosken RA, Grobler D, Van Altena JJ, Bertschinger HJ & Kirkpatrick JF 2000 Immunocontraception of African elephants. *Nature* **407** 149.
- Finkel E 2001 Australia. Engineered mouse virus spurs bioweapon fears. *Science* **291** 585.
- Fleming PL, Corbett L, Harden R & Thomson P 2001 *Managing the Impacts of Dingoes and Other Wild Dogs*, Canberra: Bureau of Rural Sciences.
- Fraker MA, Brown RG, Gaunt GE, Kerr JA & Pohajdak B 2002 Long-lasting, single-dose immunocontraception of feral fallow deer in British Columbia. *Journal of Wildlife Management* **66** 1141–1147.
- Freeland WJ & Choquenot D 1990 Determinants of herbivore carrying capacity: plants, nutrients and *Equus asinus* in Northern Australia. *Ecology* **71** 589–597.
- Furbish CE & Albano M 1994 Selective herbivory and plant community structure in a mid-Atlantic salt marsh. *Ecology* **75** 1015–1022.
- Gorman SF, Levy JK, Hampton AL, Collante WR, Harris AL & Brown RG 2002 Evaluation of a porcine zona pellucida vaccine for the immunocontraception of domestic kittens (*Felis catus*). *Theriogenology* **58** 135–149.
- Grandy JW & Rutberg AT 2002 An animal welfare view of wildlife contraception. *Reproduction (Cambridge, England) Supplement* **60** 1–7.
- Gupta SK & Talwar GP 1994 Contraceptive vaccines. *Advances in Contraceptive Delivery Systems* **10** 255–265.
- Hanks J 2001 Conservation strategies for Africa's large mammals. *Reproduction, Fertility, and Development* **13** 459–468.
- Hardy CM & Mobbs KJ 1999 Expression of recombinant mouse sperm protein sp56 and assessment of its potential for use as an antigen in an immunocontraceptive vaccine. *Molecular Reproduction and Development* **52** 216–224.
- Hardy CM, Pekin J & ten Have JF 2002a Mouse-specific immunocontraceptive polypeptide vaccines. *Reproduction (Cambridge, England) Supplement* **60** 19–30.
- Hardy CM, ten Have JF, Mobbs KJ & Hinds LA 2002b Assessment of the immunocontraceptive effect of a zona pellucida 3 peptide antigen in wild mice. *Reproduction, Fertility, and Development* **14** 151–155.
- Harrenstien LA, Munson L, Chassy LM, Liu IK & Kirkpatrick JF 2004 Effects of porcine zona pellucida immunocontraceptives in zoo felids. *Journal of Zoo and Wildlife Medicine* **35** 271–279.
- Hill AV 2001 The genomics and genetics of human infectious disease susceptibility. *Annual Review of Genomics and Human Genetics* **2** 373–400.
- Ivanova M, Petrov M, Klissourska D & Mollova M 1995 Contraceptive potential of porcine zona pellucida in cats. *Theriogenology* **43** 969–981.
- Jackson RJ, Ramsay AJ, Christensen CD, Beaton S, Hall DF & Ramshaw IA 2001 Expression of mouse interleukin-4 by a recombinant ectromelia virus suppresses cytolytic lymphocyte responses and overcomes genetic resistance to mousepox. *Journal of Virology* **75** 1205–1210.
- Joshi SH 1973 An immunological approach to fertility control. *American Journal of Pharmacy* **145** 22–26.
- Kerr PJ, Jackson RJ, Robinson AJ, Swan J, Silvers L, French N, Clarke H, Hall DF & Holland MK 1999 Infertility in female rabbits (*Oryctolagus cuniculus*) alloimmunized with the rabbit zona pellucida protein ZPB either as a purified recombinant protein or expressed by recombinant myxoma virus. *Biology of Reproduction* **61** 606–613.
- Kirkpatrick JF, Liu IKM, Turner JW & Bernoco M 1991 Antigen recognition in feral mares previously immunized with porcine zonae pellucidae. *Journal of Reproduction and Fertility. Supplement* **44** 321–325.
- Kirkpatrick JF, Naugle R, Liu IKM & Turner JW 1995 *Effects of Seven Consecutive Years of Porcine Zonae Pellucidae Contraception on Ovarian Function in Feral Mares*, *Biology of Reproduction Monograph Series*, vol 1, pp 411–413 (Equine Reproduction VI).
- Kitchener AL, Edds LM, Molinia FC & Kay DJ 2002 Porcine zonae pellucidae immunization of tammar wallabies (*Macropus eugenii*): fertility and immune responses. *Reproduction, Fertility, and Development* **14** 215–223.
- Klein J, Satta Y, O'Huigin C & Takahata N 1993 The molecular descent of the major histocompatibility complex. *Annual Review of Immunology* **11** 269–295.
- Kramnik I, Dietrich WF, Demant P & Bloom BR 2000 Genetic control of resistance to experimental infection with virulent *Mycobacterium tuberculosis*. *PNAS* **97** 8560–8565.

- Krebs JR, Anderson RM, Clutton-Brock T, Donnelly CA, Frost S, Morrison WI, Woodroffe R & Young D 1998 Badgers and bovine TB: conflicts between conservation and health. *Science* **279** 817–818.
- Lawson M 1995 Rabbit virus threatens ecology after leaping the fence. *Nature* **378** 531.
- Leenaars PP, Hendriksen CF, Angulo AF, Koedam MA & Claassen E 1994 Evaluation of several adjuvants as alternatives to the use of Freund's adjuvant in rabbits. *Veterinary Immunology and Immunopathology* **40** 225–241.
- Leenaars PP, Koedam MA, Wester PW, Baumans V, Claassen E & Hendriksen CF 1998 Assessment of side effects induced by injection of different adjuvant/antigen combinations in rabbits and mice. *Laboratory Animals* **32** 387–406.
- Lively CM & Dybdahl MF 2000 Parasite adaptation to locally common host genotypes. *Nature* **405** 679–681.
- Mackintosh CG, Qureshi T, Waldrup K, Labes RE, Dodds KG & Griffin JF 2000 Genetic resistance to experimental infection with *Mycobacterium bovis* in red deer (*Cervus elaphus*). *Infection and Immunity* **68** 1620–1625.
- Mahlow JC & Slater MR 1996 Current issues in the control of stray and feral cats. *Journal of the American Veterinary Medical Association* **209** 2016–2020.
- Martin R & Handasyde K 1999 *The Koala: Natural History, Conservation and Management*, Sydney: University of New South Wales Press Ltd.
- McCool CJ 1981 *Feral Donkeys in the Northern Territory: Report to the Feral Animals Committee of a Working Party on the Feral Donkey Problem*, Darwin: Department of Primary Production.
- McKenzie JA 1996 *Ecological and Evolutionary Aspects of Insecticide Resistance*, Texas, USA: Academic Press/R.G. Landes.
- McNicholl JM & Cuenco KT 1999 Host genes and infectious diseases. HIV, other pathogens, and a public health perspective. *American Journal of Preventive Medicine* **16** 141–154.
- McShea WJ, Underwood HB & Rappole JH 1997 *The Science of Overabundance: Deer Ecology and Population Management*, Washington, D.C.: Smithsonian Institution Press.
- Miller LA, Johns BE, Elias DJ & Crane KA 1997 Comparative efficacy of two immunocontraceptive vaccines. *Vaccine* **15** 1858–1862.
- Miller LA, Johns BE & Killian GJ 2000a Immunocontraception of white-tailed deer using native and recombinant zona pellucida vaccines. *Animal Reproduction Science* **63** 187–195.
- Miller LA, Johns BE & Killian GJ 2000b Immunocontraception of white-tailed deer with GnRH vaccine. *American Journal of Reproductive Immunology* **44** 266–274.
- Miller LA, Johns BE & Killian GJ 2000c Long-term effects of PZP immunization on reproduction in white-tailed deer. *Vaccine* **18** 568–574.
- Mohn R & Bowen WD 1996 Grey seal predation on the Eastern Scotian Shelf: modeling the impact on Atlantic cod. *Canadian Journal of Fisheries and Aquatic Sciences* **53** 2722–2738.
- Montague TLe 2000 *The Brushtail Possum: Biology, Impact and Management of an Introduced Marsupial*, Lincoln, NZ: Manaaki Whenua Press.
- Moore HD, Jenkins NM & Wong C 1997 Immunocontraception in rodents: a review of the development of a sperm-based immunocontraceptive vaccine for the grey squirrel (*Sciurus carolinensis*). *Reproduction, Fertility, and Development* **9** 125–129.
- Naz RK & Zhu X 1998 Recombinant fertilization antigen-1 causes a contraceptive effect in actively immunized mice. *Biology of Reproduction* **59** 1095–1100.
- Newsome AE 1991 Feral cats: an overview. In *The Impact of Cats on Native Wildlife*, Ed. C Potter. Canberra: Australian National Parks and Wildlife Service.
- North RJ & Medina E 1998 How important is Nramp1 in tuberculosis?. *Trends in Microbiology* **6** 441–443.
- Nowak MA & May RM 1994 Superinfection and the evolution of parasite virulence. *Proceedings of the Royal Society of London. Series B* **255** 81–89.
- O'Hern PA, Liang ZG, Bambra CS & Goldberg E 1997 Colinear synthesis of an antigen-specific B-cell epitope with a 'promiscuous' tetanus toxin T-cell epitope: a synthetic peptide immunocontraceptive. *Vaccine* **15** 1761–1766.
- Oogjes G 1997 Ethical aspects and dilemmas of fertility control of unwanted wildlife: an animal welfarist's perspective. *Reproduction, Fertility, and Development* **9** 163–167.
- Palmer MV, Waters WR & Whipple DL 2002 Milk containing *Mycobacterium bovis* as a source of infection for white-tailed deer fawns (*Odocoileus virginianus*). *Tuberculosis (Edinburgh, Scotland)* **82** 161–165.
- Pimm SL & van Aarde RJ 2001 Population control: African elephants and contraception. *Nature* **411** 766.
- Pople T & Grigg G 1999 *Commercial Harvesting of Kangaroos in Australia*, Canberra: Environment Australia.
- Rao AJ 2001 Is there a role for contraceptive vaccines in fertility control?. *Journal of Biosciences* **26** 425–427.
- Richter J 1994 Anti-fertility 'vaccines': a plea for an open debate on the prospects of research. *Newsletter (Women's Global Network on Reproductive Rights)* **46** 3–5.
- Sabeta CT, Bingham J & Nel LH 2003 Molecular epidemiology of canid rabies in Zimbabwe and South Africa. *Virus Research* **91** 203–211.
- Saunders GR, Coman B, Kinnear J & Braysher M 1995 *Managing Vertebrate Pests: Foxes*, Canberra: Bureau of Rural Science.
- Seamark RF 2001 Biotech prospects for the control of introduced mammals in Australia. *Reproduction, Fertility, and Development* **13** 705–711.
- Shideler SE, Stoops MA, Gee NA, Howell JA & Lasley BL 2002 Use of porcine zona pellucida (PZP) vaccine as a contraceptive agent in free-ranging tule elk (*Cervus elaphus nannodes*). *Reproduction (Cambridge, England) Supplement* **60** 169–176.
- Stevens VC 1975 Potential control of fertility in women by immunization with HCG. *Research in Reproduction* 1–2.
- Turner A & Kirkpatrick JF 2002 Effects of immunocontraception on population, longevity and body condition in wild mares (*Equus caballus*). *Reproduction (Cambridge, England) Supplement* **60** 187–195.
- Turner JW, Liu IKM & Kirkpatrick JF 1996 Remotely delivered immunocontraception in free-roaming feral burros (*Equus asinus*). *Journal of Reproduction and Fertility* **107** 31–35.
- Turner JW, Liu IKM, Rutberg AT & Kirkpatrick JF 1997 Immunocontraception limits foal production in free-roaming feral horses in Nevada. *Journal of Wildlife Management* **107** 31–35.
- Turner JW, Liu IKM, Flanagan DR, Bynum KS & Rutberg AT 2002 Porcine zona pellucida (PZP) immunocontraception of wild horses (*Equus caballus*) in Nevada: a 10 year study. *Reproduction (Cambridge, England) Supplement* **60** 177–186.
- Tyndale-Biscoe CH 1991 Fertility control in wildlife. *Reproduction, Fertility, and Development* **3** 339–343.
- Tyndale-Biscoe CH 1994 Virus-vectored immunocontraception of feral mammals. *Reproduction, Fertility, and Development* **6** 281–287.
- Warren RJ 1997 The challenge of deer overabundance in the 21st century. *Wildlife Society Bulletin* **25** 213–214.
- Williams K, Parer I, Coman B, Burley J & Braysher M 1995 *Managing Vertebrate Pests: Rabbits*, Canberra: Bureau of Rural Science.
- Woolhouse ME, Webster JP, Domingo E, Charlesworth B & Levin BR 2002 Biological and biomedical implications of the co-evolution of pathogens and their hosts. *Nature Genetics* **32** 569–577.
- Zenger KR, McKenzie LM & Cooper DW 2002 The first comprehensive genetic linkage map of a marsupial: the tammar wallaby (*Macropus eugenii*). *Genetics* **162** 321–330.

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