World Society for the Protection of Animals

COMPANION ANIMALS UNIT

Non-surgical methods for controlling the reproduction of dogs and cats

Internal document: guidance for WSPA staff and member societies

Aim: This document is intended to provide a brief overview of current knowledge regarding the practical application of non-surgical contraception and sterilisation for dogs and cats. This will hopefully be useful for WSPA staff and member society, who might be interested in the application of such agents for population control measures during the course of their work. It is anticipated that this document will be updated regularly as new research appears in the literature.

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Introduction

Research into the non-surgical control of reproduction in animals has focused on population management in wildlife and companion animals (owned, stray dogs and cats; and feral horses). However, many of the products that are currently available for commercial use in companion animals are aimed at owned animals. These products lead to temporary and reversible changes in fertility (contraception) so that owners have more control over when an animal can breed (Table 1). These commercial products are unlikely to be suitable for the mass sterilisation (the permanent/irreversible loss of fertility) campaigns that may be required as part of a comprehensive set of measures for the management of dog and cat populations. A non-surgical contraceptive or sterilisation technique to control breeding in stray animal populations will have different requirements than those already developed for the owned companion animal population.

The following list outlines desirable characteristics for chemical sterilisation methods specific to stray and feral companion animal populations:

- causes permanent loss of fertility;
- causes permanent loss of sexual behaviour (therefore reduces 'nuisance' behaviour of animals for people in the community and might decrease displays of some forms of aggressive behaviour);
- is effective in dogs and cats, males and females;
- requires single practical delivery (oral delivery by injection or via bait would be most practical);
- is safe and has no deleterious side effects for the target and non-target species (including humans) in case of accidental exposure or self injection;
- has good efficacy (high success rate in treated animals);
- is technically feasible;
- is stable in formulation, to allow for storage and handling under field conditions;
- allows large-scale manufacturing;
- is affordable and cost effective.

In addition, the use of non-surgical methods of sterilisation might be beneficial in situations where animal owners have specific objections relating to the neutering of animals. Some examples of common objections are:

- surgery will be painful and places the animal at risk because it requires general anaesthesia;
- surgical removal of the ovaries, uterus, or testes is unnatural and objectionable.

A recent study conducted in Brazil explored the main reasons for the avoidance of surgical sterilisation of adopted shelter dogs; reasons cited included compassion (56.5%) and believing the procedure is unnecessary (11.4%)[1].

Main methods for the non-surgical control of reproduction in animals

Immunocontraception

Includes: GonaCon™ (National Wildlife Research Centre, USDA), Canine Gonadotrophin Immunotherapeutic Factor (Pfizer Animal Health), SpayVac™ (Spay Vac for Wildlife Inc.).

This approach to control reproduction uses the body's own immune system to inhibit fertility. Certain methods may also be referred to as immunosterilisation or immunocastration. Introduction of exogenous reproductive proteins (antigens) via injection triggers the animal to produce antibodies, which also act against its own (endogenous) reproductive hormones and proteins; neutralising their activity and inhibiting the normal reproductive processes.

Several biological targets have been selected for immunocontraceptives including:

1. The zona pellucida (ZP). This is the coating on the oocyte (egg), which the sperm binds to during fertilisation. Porcine ZP antigens stimulate female mammals to produce antibodies that adhere to the surface of the eggs, preventing sperm from binding and therefore blocking fertilisation.

2. Reproductive hormones (testosterone/oestrogen) through the inhibition of gonadotrophin releasing hormone (GnRH). GnRH stimulates synthesis and secretion of luteinizing hormone (LH) and follicle stimulating hormone (FSH), which are both secreted from the anterior pituitary gland and determine testicular and ovarian function through feedback mechanisms effecting the secretion of oestrogen and testosterone. (See Table 1)

The main advantage of immunocontraception is that it might be suitable for oral administration[2], which could be delivered via a bait and hence avoid the need for animal capture. Vaccines against GnRH also have the advantage (for stray populations) of suppressing sexual behaviour in males and females[3].

The availability of these products is increasing. One GnRH vaccine, available in the USA (a canine gonadotrophin releasing factor immunotherapeutic), is marketed for the treatment of a medical condition resulting from age related hormone alterations in the dogs' prostrate gland: Benign Prostatic Hypertrophy (BPH) rather than contraception. Another vaccine (GonaCon™) is currently being investigated for use in dogs and cats.

Further challenges must be overcome before these are suitable for large scale injectable or oral use with free-ranging populations. The main concerns at this stage are unquantified side effects, achieving efficacy over long periods of time following single injection (currently boosters seem to be required to ensure continued efficacy) and either species-specific effect or delivery in the case of oral baits, to ensure infertility does not spread beyond the target population.

Hormonal down-regulation

Includes Suprelorelin® (Peptech Animal Health), Gonazon® (Intervet), Ovaban (Schering-Plough Animal Health), Delvosteron® (Intervet).

The use of synthetic (exogenous) steroid hormones suppresses fertility by inhibiting production of endogenous hormones (down-regulation). This method has been used extensively for contraception in human and non-human animals.

Both synthetic progestins (Megestrol acetate, Medroxyprogesteron acetate (MPA) and proligestone) and androgens (Mibolerone and Danazol) are used in veterinary medicine for the control of conditions exacerbated or caused by steroid sex hormones; and for the management of behavioural problems that might be under the influence of testosterone or oestrogen. These are only suitable for short-term use and can be administered, orally or by injection, at daily or weekly intervals by owners (when suitable). The factor preventing these hormones being used for stray animal contraception is the need for regular dosing over a protracted time period. This is both impractical and associated with adverse effects if used long-term. In addition, without repeated dosing reproductive capacity could rapidly resume.

GnRH agonists (Deslorelin, Suprelorelin®, Gonazon®) have been developed for use as contraceptives in male and female dogs[4]. These agonists are administered via implants and are reversible. They reduce the need for frequent dosing as the active chemical is slowly released from the implant. However, implants need to be replaced regularly (Suprelorelin® every 6 months; Gonazon® every 12 months) to maintain infertility.

The long-term side effects of progestins and androgens (see Table 1) make them unsuitable for use in stray or feral populations. GnRH agonists have been used successfully in wildlife, although the side effect of induced oestrus has not been eliminated. In addition, GnRH agonists currently rely on being administered to dogs before their first oestrus.

Intratesticular, intraepididymal and intra-vas-deferens injections

Includes: Neutersol® (Abbott Laboratories).

More commonly known as chemical castration, this method causes permanent infertility in males treated at a young age by inducing azoospermia (no measurable level of sperm in the semen). The method requires injection directly into the testicles (two injections; one in each testicle), which creates some discomfort and requires the animal to be appropriately restrained. Sedation may or may not be used for dogs and anaesthesia used for cats, although this is suggested by the manufacturer to facilitate handling rather than to mask any discomfort from the procedure. The manufacturer claims that administration of these injections is not painful.

Neutersol®, a cytotoxic substance registered for use in the USA is a zinc gluconate solution neutralised by arginine. When injected directly into the testicle Neutersol® causes atrophy of the testes and prostrate gland resulting in permanent sterilisation. In the USA it is currently approved for use in dogs aged between 3 and 10 months although it is being tested for use in older animals. In Thailand this is being trialled in adult animals without prior sedation. Although Neutersol® can be injected without sedation, the dog must be held firmly on its back to enable accurate delivery and it would be useful to facilitate restraint by sedation. The procedure requires a degree of skill: if injected outside the testicle (during insertion or withdrawal) the substance can be highly irritating and lead to ulceration of the tissues.

Neutersol® application, although associated with a 41–52% reduction in testosterone levels (post dose), might not alter sexually dimorphic behaviour in treated animals. Roaming, marking, aggression and mounting, for example, might still be displayed. In clinical trials researchers reported Neutersol® to be 99.6% effective when administered according to the manufacturer's instructions. Adverse reactions observed during clinical trials include swelling of the testes, scrotal pain when palpated (6.3%), anorexia (4.1%), diarrhoea (2%) and lethargy (2.2%). Severe scrotal ulceration was found in 4% of dogs, 2% of which required surgical intervention including scrotal ablation and removal of the affected necrotic tissue – a more complicated surgical procedure in comparison to routine castration surgery. Depending upon the skill of the veterinarian and the accessibility of suitable anaesthetics and analgesics, the side effects reported in this minority could eventually lead to the euthanasia of those dogs affected.

Products in development for intra-testicular injection which result in azoosperma include calcium chloride[6] and a novel zinc-based solution[7]. Both have been shown to be effective under experimental conditions but are in the early stages of clinical development.

Chemical targeting

Includes: ChemSpay.

This is a novel methodology using a chemical (industrial toxicant) that specifically targets the ovary. Chemical targeting causes the depletion of the primordial and primary follicles leading to permanent sterilisation and elimination of oestrus. This method is at the very early stages of development by researchers in the USA. At present, sterilisation can only be achieved by a series of injections, although researchers are trying to develop an effective single dose delivery through injection or oral dose delivered via bait. However, care and evidence of injection safety would be required to ensure human safety for female (operators) as this is not a species-specific active ingredient.

Cytotoxin conjugates

Includes: GnRH analogue linked to PAP.

Cytotoxin conjugates are plant toxins linked to a GnRH analogue that when injected into the body bind to gonadotrophin releasing cells in the pituitary gland. The plant toxins destroy these cells in the pituitary gland and therefore inhibit the release of luteinising hormone (LH) and follicle stimulating hormone (FSH).

This method is currently being researched at Colorado State University by Terry Nett[8]. Researchers have conjugated a GnRH analogue to pokeweed antiviral protein (PAP). PAP is a ribosome inhibiting protein, highly toxic but difficult to introduce into cells. In trials it is reported to cause a dramatic reduction in serum and pituitary LH (>90%). Renal toxicity has been observed when the compound is given at high doses; this is assumed to result from free PAP that is not conjugated to a GnRH analogue. No such side effects are observed when the compound is given at lower doses. Current research indicates that the desired effect is not reached until 4 weeks after dosing. Some early clinical tests in dogs indicate that the technique has been successful in causing infertility, and current research suggests that this effect is permanent when delivered into adult animals. However, this might not be the case when given to pre-pubertal animals, as the pituitary gland might still be developing and new cells that release gonadotrophin could grow sometime after the compound has been excreted from the body. The researchers have yet to complete long term clinical trials; nevertheless this technique might well be one to watch out for in the future.

Other methods

Includes: mechanical.

Several other methods have been used as contraception for dogs and cats without much success. Mechanical barriers and intrauterine devices, for instance, have a high failure rate and are difficult to fit.

Mechanical sterilisation using ultrasound is effective at causing sterility in male dogs, but must either be used at a low level, which requires repeated applications, or at a higher level, which has a high (20%) chance of skin burns[9]. These methods were developed in dogs under anaesthesia, to facilitate handling for the desired length of time and to ameliorate the aversive nature of this method. This would render them impractical for use in field conditions.

Summary

The development and application of a suitable, effective, non-surgical method of permanent sterilisation of animals would have an enormous advantage over the current surgical procedure because animals could be treated without being anaesthetised. At present the most promising methods are the development of immunocontraception/sterilisation vaccines or cytotoxin conjugates because both can be delivered as a single dose.

Other methods currently rely on repeated dosing for long-term suppression of reproduction; therefore their application to stray animal population control is limited.

The table on pages 9 to 12 summarises the essential characteristics of each method.

Table 1. Current chemical contraception and sterilisation products available and undergoing clinical trials

PRODUCT	STATUS	TYPE	MODE OF ACTION	TREATMENT	DURATION	TARGET ANIMALS	CONTRAINDICATIONS/ COMMENTS	APPLICATION
ChemSpay® Produced by: SenesTech www.senestech.com	Early development Trials in the USA	**(IV)** Chemical highly specific to the ovary	Chemical selectively depletes primordial and primary follicles in the ovary Total irreversible ovarian failure Permanent sterilisation Eliminates oestrus and oestrus behaviour	Early stage of development Currently a series of injections Aim to develop a single injection and oral dose for use in bait	Early stage of development Permanent	Dogs Cats *Females*	Non reported Early stage of development Clinical trials being conducted	Has potential for use in female dogs and cats as part of a population management strategy Female operator safety is a concern
Suprelorelin® Produced by: Peptech Animal Health www.peptech.com/HTML/Animal_Health/AnimalHealth.html	Available AU, NZ Approved EU US approval being sought	**(II)** GnRH agonist	Slow release of the active product from the implant Halts production and release of LH and FSH Reduced testosterone production and circulating levels in the blood Halts sperm production	Cylindrical implant similar size to microchip Subcutaneous injection Implant inserted just under the skin between the shoulder blades Does not require an anaesthetic	6 or 12 months In clinical trials – 5 consecutive treatments - suppressed reproductive function in male dogs for 3 years Fertility returns post final treatment	Dogs *Males*	Short-term suppression of reproductive function Requires repeat doses Testosterone levels decline – might affect sexually dimorphic behaviour influenced by testosterone None reported from the trial data. Not licensed for long term use? Should not be given to pregnant females High cost per treatment	Owned male dogs Temporary, reversible suppression of reproductive function in male dogs Repeat dosing and cost of application make it impractical for use in a comprehensive population management strategy
Neutersol® Produced by: Abbott Laboratories www.abbott.com	Approved USA Trials in Thailand, India and Mexico	**(III)** Zinc Gluconate (+ Arginine)	Cytotoxic Causes atrophy of the testes and prostrate gland Halts sperm production Reduced testosterone production and circulating levels in the blood	Intratesticular injection Precise injection into the testes, behind the epididymis	Permanent if used in young male dogs. Impact on adult dogs unkown.	Dogs *Males* Aged 3-10 months Future use in *male* cats	Permanent cessation reproductive capacity Mexico study: 10,000 dogs underwent the procedure; reported 97% effective (not adult dogs) Must avoid injection into scrotal sack and skin Requires manual restraint Testosterone levels decline: might affect sexually dimorphic behaviour influenced by testosterone – although this is not reported Not suitable for use in dogs that are: Cryptorchid Pre-existing scrotal irritation or dermatitis Diseased or malformed testes Testicular width < 10mm or >27mm	Welfare concerns remain a major barrier for its use outside closely supervised dogs with access to good veterinary care, as side effects can be significant in a small proportion of treated animals.

PRODUCT	STATUS	TYPE	MODE OF ACTION	TREATMENT	DURATION	TARGET ANIMALS	CONTRAINDICATIONS/ COMMENTS	APPLICATION
							Swelling 2- 7 days post injection Severe reactions result in ulceration of testicles and scrotum requiring surgical intervention and possible euthanasia Pain and irritation often not assessed when dogs are released Irritation may lead to licking and self trauma	
Gonazon® Produced by: Intervet* France www.intervet.com	Approved EU (2006)	**(II)** GnRH agonist	Long term blockade of gonadtrophin synthesis in bitches Prevents ovulation Eliminates oestrus and oestrus behaviour	Rectangle implant, dimensions: 14 x 3 x 1 mm Subcutaneous injection Implant inserted in the region of the umbilicus	1- 2 years Reversible, on removal of the implant	Licensed for Dogs *Females* Aged 4 months - 6 years Future use: Dogs Cats *Males* *Females*	Longer term suppression of reproductive behaviour Requires repeat treatment? Expensive to produce but might last longer if implant not removed Oestrus behaviour is not displayed Clinical trails report rare cases of vaginitis in pre-pubertal females	Owned female dogs Cost might be prohibitive and might require removal of implant and re-implantation if effect is to be permanent Unlikely to be suitable for use as part of a population management strategy
GonaCon™ Produced by: National Wildlife Research Centre, USDA – wildlife services www.aphis.usda.gov/ws/nwrc/research/reproductive_contol/gonacon.html	Clinical trails conducted in *Cervids* in USA Due for approval by Environment Protection Agency (EPA) in USA for white tailed deer Suggested development for application in dogs and cats	(I) GnRH vaccine	Induces the body to make antibodies against its own GnRH Stops the production of sex hormones oestrogen/testosterone Promotes infertility in both males and females May alter sexually dimorphic behaviour	Single injection Intramuscular injection	Suppression of reproduction for up to 2.5 years in field trials	Cervids *Males* *Females* Future use: Dogs Cats *Males* *Females*	Long-term suppression of reproductive behaviour Sexually dimorphic behaviour under the influence of oestrogen or testosterone might be reduced When trialled in Elk 2/10 animals reported to have injection site reactions; even up to 18 – 20 months post dose Injection site reaction also reported in dogs; minority so severe that euthanasia needed (appears to be related to the adjuvant – so possibilities to use different one or lower dose) Injection site reactions not reported in cats?	Requires animals to be caught and injected Could be suitable for application for stray animals in injectable or oral vaccine form in the near future – most likely as annual vaccine delivered at same time as rabies vaccine. Reactions sites low prevalence/high severity in dogs at present make it currently undesirable. However, the problem of injection site reactions is being explored and could be overcome.

PRODUCT	STATUS	TYPE	MODE OF ACTION	TREATMENT	DURATION	TARGET ANIMALS	CONTRAINDICATIONS/ COMMENTS	APPLICATION
Canine Gonadotropin Releasing Factor Immuno-therapeutic Produced by: Pfizer Animal Health www.pfizerah.com	Available USA	**(I)** GnRH vaccine	Treatment of BPH in entire male dogs Analogue of GnRH linked to a carrier protein Induces the body to make antibodies against its own GnRH Stops the production of testosterone Promotes infertility in males Might alter sexually dimorphic behaviour	Injection Primary vaccination requires 2 doses given 4 -6 weeks apart Repeated vaccine interval is 6 months	6 months	Dogs *Males* (entire) Post-puberty	Not permanent or long term Might affect sexually dimorphic behaviour under the influence of testosterone No systemic or adverse reactions reported within 14 days of administration during clinic trials	Currently licensed for use in dogs with BPH Might be suitable for owned male dogs Because repeated vaccination is required at frequent intervals (6 months) it is unsuitable for use in comprehensive population management programmes
Ovaban Produced by: Schering – Plough Animal Health www.spah.com	Available USA and EU	**(II)** Megestrol acetate Progestogen	Postponement of oestrus Treatment of false pregnancy	Oral Tablet to be given Daily dosing for prescribed time	Daily dosing required: Dependent upon stage of bitches oestrus cycle 8 days dosing required for dogs in pro-oestrus	Dogs *Females* Post-puberty	Short-term postponement of oestrus Not to be used for postponing first oestrus Can take 3 – 8 days before signs of oestrus (vaginal bleeding and vulva swelling) disappear During this time bitches might still accept male dogs Females should be separated from male dogs until signs of oestrus have subsided Side effects in prolonged treatment: Sustained over dosing associated with cystic endometrial hyperplasia in clinic trails Transient effects reported in clinic trials: Increased appetite Changes in temperament Enlarged mammary glands Lactation Pyometra (0.6% of cases) Sustained use of megastrol acetate is associated with mammary tumours, uterine lesions, pyometra	Owned female dogs Temporary postponement of oestrus only or treatment of false pregnancy Not suitable for use in comprehensive population management programmes

PRODUCT	STATUS	TYPE	MODE OF ACTION	TREATMENT	DURATION	TARGET ANIMALS	CONTRAINDICATIONS/ COMMENTS	APPLICATION
Delvosteron® Produced by: Intervet* www.intervet.com	Available NZ, UK, and many other countries	**(II)** Progestagen	Control of oestrus in female animals Permanent or temporary postponement of oestrus can be achieved	Injection Subcutaneous	Initial, repeated 3 months and 7 months later Permanent postponement of oestrus achieved with repeated injections given in anoestrus/ metoestrus Suppression of oestrus requires injection to be given at the beginning of proestrus Temporary postponement is achieved with a single injection given when in anoestrus	Dogs Cats *Females*	Mainly used for short-term suppression of breeding Advised to use after first oestrus Doesn't immediately stop signs of oestrus after administration Advised to separate females from males for up to 5 days after administration Side effects associated with use of progestagens: Can cause adrenal suppression in some animals Cystic endometrial hyperplasia/pyometra	Owned animals Repeated treatments required at frequent intervals and at specific times during oestrus Unlikely to be applicable to comprehensive population management programmes Side effects associated with long-term use
SpayVac™ Produced by: SpayVac™-for-wildlife, Inc. www.spayvac.org	Efficacy & safety trials conducted in USA 4 – 5 years before FDA approval for use in USA	(I) Zona pellucida vaccine	Blocks fertilisation Porcine Zona Pellucida antigens stimulate female mammals to produce antibodies that adhere to the surface of her eggs Prevents sperm from binding and therefore blocks fertilisation	Injection	Single dose has been found to prevent reproduction for up to 3 years in female Deer	Cervids *Females* Future use: Dogs Cats *Females*	Long term but not permanent No reported side effects in field trials in wildlife Some reported effects in horses – fractious or irritable behaviour Wouldn't expect changes in oestrus and oestrus related behaviour	Lack of impact on reproductive behaviour make this undesirable for application to population management programmes Current development requires capture and handling for injection – suitable for oral delivery in baits in the future?

Key: (I) Immunocontraception /immunosterilisation (II) Hormonal down-regulation (III) Intra-testicular injection (IV) Chemical
***Intervet has subsequently been taken over by Schering-Plough Animal Health**

References

1. Soto, FRM; Ferreira, F; Pinheiro, SR; Nogari, F; Risseto, MR; de Souza, O; Amaku, M. (2005) Adoption of shelter dogs in a Brazilian community: assessing the caretaker profile. *Journal of Applied Animal Welfare Science* 8:105-116

2. Purswell BJ, Kolster KA. (2006) Immunocontraception in companion animals. *Theriogenology* 66:510-513.

3. Kutzler M and Wood A. (2006) Non-surgical methods of contraception and sterilization. *Theriogenology* 66:514-525.

4. Rubion S, Desmoulin PO, Riviere-Godet E, Kinziger M, Salavert F, Rutten F, Flochlay-Sigognault A and Driancourt MA. (2006) Treatment with a subcutaneous GnRH agonist containing controlled release device reversibly prevents puberty in bitches. *Theriogenology* 66(6-7): 1651-1654.

5. From HSUS website www.animalsheltering.org

6. Jana K and Samanta PK. (2007) Sterilization of male stray dogs with a single intratesticular injection of calcium chloride: a dose-dependant study. *Contraception* 75(5): 390-400.

7. Oliveira ECS, Moura MR, Silva VA, Peixoto CA, Saraiva KLA, Cavalcanti de Sa MJ, Douglas RH and de Pinho Marques Jr. A. (2007) Intratesticular injection of a zinc-based solution as a contraceptive for dogs. *Theriogenology* 68: 137-145.

8. Sixth International Conference on Fertility Control for Wildlife. York, 3rd – 5th September 2007.

9. Roberts WW, Chan DY, Fried NM, Wright EJ, Nicol T, Jarrett TW, Kavoussi LR, Solomon SB. (2002) High intensity focused ultrasound ablation of the vas deferens in a canine model. *Journal of Urology* 167:2613-2617.

10. Gobello C. (2007) New GnRH analogs in canine reproduction. *Animal Reproduction Science* 100(1-2): 1-13.

11. *The Alliance for Contraception in Cats and Dogs website. (www.acc-d.org). Contains 2006 proceedings from The Third International Symposium on Non-surgical Contraceptive Methods of Population Control.*