MALE INFERTILITY AND TREATMENTS IN THE CANINE SPECIES

J.VERSTEGEN, DVM, MSC, PHD, DECAR, K. ONCLIN, DVM, PHD, DECAR
VETERINARY COLLEGE OF THE UNIVERSITY OF LIÈGE DEPARTMENT OF ANIMAL CLINICAL SCIENCES, SMALL ANIMAL THERIOGENOLOGY.
Bd Colonster 20 B44, B4000 Liège Belgium email: J.Verstegen@ulg.ac.be

Infertility in the male dog and cat is not a current process in the whole canine and feline population. However, in our specialized everyday life clinic and activity, specialized veterinarians are, more and more often, visited for problems related to poor reproduction abilities or reduced/absent fertility in some valuable animals. Breeders are more and more led by economical targets and request more and more from their male animals. This trend to higher productivity is often linked to fertility problems. Furthermore, they are consistently aware of the crucial role the veterinarian may nowadays play and the availability of new techniques and drugs he can provide to further improve reproduction. It is however important to emphasize the fact that, due to the specific biology of spermatogenesis and the long duration of this one, treatments, except when the origin of the problem is clearly established, are often disappointing, particularly in the fact that a long interval between the beginning of the treatment and the first clinical improvement is observed. The available tests to evaluate objectively fertility are scarce and, in general, of poor predictive value. Male infertility is a recent area in veterinary clinics and purebred dogs are mainly concerned. The etiology and treatments, like in humans, are often unclear and it is essential for the veterinarian to have a minimal knowledge of the main causes, a good way towards diagnosis and a certain skill in practicing the additional examinations including spermogram.

Definition of the problem.
History is in general essential in the clear determination of the actual problem and should certainly include informations like past illnesses, previous medications, type and quality of food, vaccinations, … as well as detailed breeding performances for not only the concerned animal, but also for all other males present in the kennel.
Male infertility varies from a complete absence of libido to the inability to sire offspring in spite of normal matings, thus history is essential to discriminate the kind of problems observed. They can go from the easiest to determine and often to cure: behavioral or anatomical defects to the more complex ones : endocrinological or infectious defects. Infertility in male dog is often proposed when whelping rate is lower than 75% of mated bitches or when more than 3 consecutively mated bitches remain empty. Veterinarians have to be aware that infertility in the dog can often be due to a wrong timing of natural or artificial insemination (50 to 80% of the cases), other problems related to the bitch and to the kennel management and finally (10 to probably less than 30%) to a real male infertility problem. In many cases, infertility is due to improper breeding management that should absolutely be carefully checked a least by evaluating usual breeding management.

Physical examination

This simple procedure allows already for the observation of acquired or inherited abnormalities of the sexual organs. The scrotum, testes (location and characteristics), the penis in normal and erected conditions as well as the prostate should be checked for determination of anatomical or morphological abnormalities.

The etiology

Infertility may be related to sexual behavior, to anatomical defects or to the semen itself. Some of the abnormalities (behavioral, anatomical or functional) can be primary or secondary defects due to other general pathologies like endocrine, infectious or metabolic diseases or more specific urogenital tract diseases (urinary or prostate diseases). Exposure to toxins, medications or idiopathic infertility may be observed in dogs like in humans.

a) Abnormal sexual behavior may include or be related to lack of libido, pain, hierarchy, breed, age, bad or poor socialization, stress, environmental temperature, …

b) Anatomical problems may be congenital: testis or deferent ducts hypoplasia, hypospadias or epispadias, micro or megapenis, persistent frenulum, unilateral or bilateral testicular ectopy (cryptorchidism), size disproportion between the male and the female, hermaphroditism,…
The acquired anatomical problems may include post traumatic or inflammatory adhesions between penis and prepuce, fracture of the bone penis, orthopedic general problems, long hair or invaginated prepuce (Afghans, Collies, Yorkshire Terriers, ..), post inflammatory fibrosis of the testis, epididymidis, prostate,…. The anatomical problems may prevent mating to occur or be responsible for azoospermia or aspermia.

c) Prostate diseases may be responsible for infertility problems by the changes in the composition and nature of the prostatic fluid, particularly in male dogs over 5-7 years where prostate pathologies are becoming frequent. If the prostatic fluid pH is over 7, asthenozoospermia can easily be observed and responsible for infertility. The presence of blood or pus in the prostatic fluid is often associated with sperm defects whereas the effects on fertility of a primary or secondary reduction of prostate fluid volume (i.e. after a finasteride or hormonal treatment) are still controversial. We have meanwhile demonstrated that pregnancy can be observed in dogs even in total absence of prostatic fluid.

d) The alcalinization of urine, often associated with bladder infection, can also be associated with asthenozoospermia.

e) Endocrine problems of central (dysfunction of the hypothalamo-pituitary axis including adenomas) or peripheral origin, including testis, thyroid or adrenal glands, may be associated with infertility. Endocrine disorders that are often associated with infertility are hypothyroidism and hyperadrenocorticism. The later, by the increase in cortisol, is associated with pituitary negative feedback on LH release and subsequent reduced secretion of testosterone by the Leydig cells.

f) Endocrine disruptors of environmental origins are, in dogs as in other species, more and more often suspected to be involved in some unexplained cases of infertility. Polluents, pesticides, chemicals (like systemic antiparasitic agents or anti-oxidants) are common in everyday dogs' life as they are in humans and other species and may also, to some extent, explain some recently observed increases in dog infertility.

g) Iatrogenic causes: some hormonal compounds (corticosteroids, anti-androgens, androgenic agents) or antifungal drugs (griseovulvine, ketoconazole), systemic antiparasitic agents are nowadays more and more known to influence reproduction.

h) Infectious diseases involving the reproductive tract often lead to infertility. This observation is well-known for Brucellosis sp. which may induce teratozoospermia with numerous neutrophils and other WBC or semen agglutination, but it could also be the case for E.coli, Proteus sp, Streptococci or Staphylococci, Pasteurella sp, Mycoplasma infections.
i) Genetic origin of infertility includes chromosomes problems, particularly in some breeds or products of line-breedings, leading from azoospermia to intersexuality (Kartegener's, XXY, XX syndromes). There is clear evidence nowadays of the influence of breed or selection on semen quality. This may be related to some influence of selection (selecting for a desired character being sometime associated with the selection of reduced fertility, like in bovine or other species)and inheritability of fertility. Some authors consider that large breeds are most affected.
j) Physical causes like local or general ischemia and hyperthermia (i.e. associated with local dermatitis) or radiations are known to be associated with anatomical and functional testis defects. The clinical significance of the later is still questionable!
k) Other causes of male infertility include: age and possible age-related andropause like in other species, auto-immune diseases often associated with semen agglutination at microscopic evaluation, organic diseases with general repercussions like kidney failure, degenerative liver diseases, diabetes mellitus, …

Diagnosis of male infertility.
The diagnosis of male infertility is based on a complete commemorative (essential initial step, involving the kennel andthe animal histories and more specifically, the previous problems related to the reproductive function), a general clinical and a specific andrological examinations. The complementary approach should include examination of reproductive behavior including libido and erection capabilities, semen collection, if possible with separation of the 3 fractions and analyzing each fraction separately, and including spermogram (table 1) bacteriological evaluation and alkaline phosphatases concentration determination, hormonal assays as well as serological tests, sonographic and radiographic examinations and finally testicular biopsy. The description of these diagnostic methodologies being behind the scope of this presentation, the readers are send to the bibliographic list available to get detailed description of the technologies.

Parameter	Description
Oligozoospermia	<200 million sperm in the entire semen-rich fraction
Teratozoospermia	<70% sperm cells with normal morphology
Asthenozoospermia	<50% progressively forward motility
Leukozoospermia	>2000 WBC per µl in the ejaculate
Azoospermia	No sperm in the ejaculate Either of testis origin: no semen produced : AP increased in the testis fraction of the ejaculate (>5000U/L) Or related to absence (or inability) of ejaculation: no increase of AP in the ejaculate
Hemospermia	Blood (grossly or at cytology) in the sperm rich fraction

Testosterone assays are of poor predictive value in absence of stimulation tests, the only ones able to guarantee the evaluation of normal testis function. Stimulation tests can be realized either using GnRH (indirect evaluation of testis function – direct evaluation of pituitary function) or

using hCG (mainly LH activity in dogs, directly evaluating testis function). If GnRH is used, 0.5µg/kg IM or 0.1µg/kg IV could be injected and the concentration of testosterone evaluated with the analyses of samples taken prior administration and 1 hour after. If hCG is used, doses of 50 UI/kg are recommended. It is however important to note that all these dosages are mainly empirical and that no real dose-response studies have been realised in dogs. In a normal male, plasma testosterone concentration is supposed to increase from 1-6 ng/ml before up to 5-12 ng/ml after stimulation. The gonadotrophins can be evaluated if the assays are available. FSH has been correlated with infertility (decline of spermatogenesis ➔ decline in inhibin production ➔ increased FSH), but without, up-to-now, convincing data ; LH concentration after GnRH stimulation allows for the determination of pituitary primary troubles. LH plasma concentration is normally lower than 20 ng/ml before and should increase rapidly over 30 ng/ml after GnRH stimulation.
LH concentration is always over 30 ng/ml in castrated animals.

Sonography of the testis is of great help as a diagnostic tool for testis tumors as well as for the detection of secretory azoospermia (no increase of AP in the ejaculate and particular aspect of the testis) whereas sonography of the epidydimis is of use in the diagnosis of excretory azoospermia either from fibrotic reactive and obstructive or tumoral origins.

Testicular biopsy is indicated in dogs that are persistently azoospermic or severely oligospermic. Incision or punch biopsies are generally preferred over percutaneous fine-needle aspiration biopsy because specimens obtained with the later often contain insufficient tubules to allow a detailed and precise diagnosis. However, even if a clear anatomopathological diagnosis is often difficult to realize, spermatogenesis can generally be assessed by the observation of sperm cells. This is a very predictive technique, but sometimes associated with some complications like hemorrhage and necrosis, infection, subsequent fibrosis and even testicular atrophy. Finally, the disruption of the blood-testis barrier may potentially induce immune-mediated orchitis.

Prognosis
The prognosis is highly dependant of the cause and in general, is good in cases of abnormal sexual behavior or secondary anatomical defects whereas it is more difficult to establish and generally worst in cases related to semen defects or following iatrogenic (toxic, therapeutic, …) origins. Leukospermia due to infection is generally of poor prognosis because of the high risk of epididymal damages, teratozoospermia is in general of poor prognosis ; however, the prognosis is most often difficult to realize. Indeed, the correlation is poor between morphological and basic functional evaluations (motility, progression, acrosome reaction) of semen and fertility. The availability of the new reproductive-assisted technologies dramatically helped improving reproduction in these animals, using uterine insemination, but raising important ethic issues! Asthenozoospermia similarly has guarded prognosis ; however again, the use of RAT (intrauterine insemination and semen dilution) may improve the prognosis. The prognosis for the other conditions will mainly depend on the ability to identify and treat the primary cause of the defect.

Treating male infertility
The recent development of reproduction-assisted technologies helped dramatically to improve treatments in many cases of male dog infertility. The treatments clearly depend on identification

of the underlying causes. Anatomical disorders have to be corrected if possible . Caution has certainly to be taken when dealing with potentially heritable problems. Disorders of sexual behavior are generally solved either by treating the behavioral troubles (hierarchy, sociabilization, disconnecting agents like low doses of diazepam to reduce the stress,...) but can also be overcome by using semen collection and artificial insemination. The aphrodisiac drugs have shown disappointing results (yohimbine 2mg/5kg/j) whereas the preliminary trials using Viagra® have been inefficacious. Disorders of erection sometimes respond to GnRH administration whereas retrograde ejaculation is said (but without clear scientific demonstration up-to-now) to be treated with the administration of phenyl-propanol-amine (PPA) known to increase urethral pressure. In dogs with poor libido or oligospermia – asthenozoospermia, the prognosis is generally poor ; however, we have got good and sometimes unexpected results with empirical GnRH treatment for 2 to 3 weeks (0.5µg/kg every 2- 3 days IM). Some androgens (like testosterone hexahydroxybenzoate 5 to 20 mg/dog/month up to 3 months or mestrolone 0.75 to 1.5 mg/kg/d for up to 3 months) have been given but with controversial results in terms of fertility. All infectious cases should be treated with high doses – long duration antibiotics like quinolones. Prostatic diseases should be treated specifically, depending on the nature of the prostate problem.
In many cases of semen related infertility, a minimum of 3 spermatogenic cycles (± 6 months) is necessary to evaluate the benefit of a treatment. It is essential to inform the owner that treating infertility of idiopathic or non clearly determined cause is always time consuming and relatively expensive for often disappointing results.

Bibliography: reference list available by contacting the author at J.Verstegen@ulg.ac.be