## Overview of canine artificial insemination for the breeder

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Insemination is the introduction of semen into the reproductive tract of a female. In mammals natural insemination follows copulation. Artificial insemination (AI) is insemination by other means. The first successful artificial insemination reported in any species was performed for a bitch in Italy in 1784. Today AI is essential in animal agriculture. It is critical for efforts to prevent species extinction, and it now is performed commonly in the pure bred dog fancy. This is an overview of items of importance when AI is considered.

Keywords: Artificial insemination, canine,

First it is important to know that for most breeds and lines the greatest number of offspring come from natural cover of a healthy female by a proven sire repeated every two days during standing heat. When it is all said and done it is hard to improve on nature.

Artificial insemination provides for conception when physical problems or circumstances prevent natural breeding of otherwise healthy and genetically sound animals. It overcomes barriers of space and time so that regardless of the location of a male his genes can be brought to a female at the precise moment necessary for fertilization of her ova. Artificial insemination combined with methods for collection, preservation, and shipping of semen can eliminate the need for travel of animals for breeding; AI can reduce risks of injury and infection of the bitch during natural breeding; and it can entirely eliminate these risks for the male.

Artificial insemination should not be used to overcome genetic problems which prevent natural breeding such as the inability to mount because of heritable joint disease. Likewise, stored semen should not be used for AI if the male has evidence of heritable disease at any time of his life even if he appeared completely healthy at the time the semen was collected.

Because AI can facilitate multiple inseminations from a single dog it can maximize the number of puppies born to a desirable male. This is a two-sided sword and can have negative consequences long term should a male be an inadvertent source of a genetic defect. This is a good reason for preserving semen from many healthy males. A breed will be better able to recover from a widespread defect if there is available stored semen from many males who are unlikely to have been affected.

When AI is done, compliance with registry requirements is important. Some registries control the breeding of all registered dogs and must give their approval before a breeding can be performed. Other registries require DNA testing for breeding performed with shipped or stored semen. Shortcuts for testing, incomplete animal and semen identification and failure to comply with registry rules can result in questioning parentage and is to be avoided always.

In a number of states artificial insemination is considered an animal husbandry task and an inseminator does not have to be a licensed veterinarian. For success and to avoid serious injury to the bitch the inseminator must understand semen processing, instrument handling, and infection control. The inseminator must have very specific knowledge of anatomy, estrous cycles, and tissue manipulation in the species they inseminate.

In the bitch the reproductive tract and surrounding tissues are complex structures which vary in size and position from breed to breed and female to female. A bitch in standing heat is remarkably tolerant of manipulation, but her tissues are delicate and easily injured. Major nerves, blood vessels and the urinary tract are close at hand. Inadvertent perforation of the vagina can occur, and insemination into the abdominal cavity rather than the vagina is life threatening.

The estrous cycle of the bitch is unique among domestic animal species. A bitch may be in season for three weeks or longer, but the time when conception can occur lasts only two days. Artificial insemination can be quite successful when it is done every other day throughout the

entire standing heat. In some bitches this may require four or more inseminations. When the number of inseminations is limited then ovulation timing to identify the fertile period is necessary. Behavior changes, vaginal swelling, vaginal discharge, and vaginal cytology can verify when a bitch is in standing heat and when she goes out, but they cannot identify ovulation and predict the fertile period. That requires blood tests for progesterone and possibly luteinizing hormone.

Semen used for AI can be freshly collected and used right away. It can be extended and chilled for short term storage or shipment. It can be frozen for many years and then thawed and used. If all other factors are equal, fresh collected semen is preferred to extended chilled semen, and extended chilled semen is preferred to frozen then thawed semen.

Collection of semen is by manual manipulation. It is best done at a location where the male feels at ease. An in-heat teaser bitch for stimulation of the male is desirable, but she is not essential if the male has been trained for collection. Semen can be collected into a variety of containers including artificial vaginas, plastic bags, and funnels with plastic tubes. Semen should be collected with clean processes and held in very clean if not sterile containers from time of collection until it is used.

Semen used for insemination must be assessed for quality when collected and at the time of insemination. Quality is determined by sperm count, sperm motility, and sperm morphology. One hundred million to two hundred million highly motile and morphologically normal sperm are minimum numbers needed for a single vaginal insemination. Conception can occur with fewer sperm with intrauterine insemination.

Fresh collected semen generally has very high total sperm numbers with good motility and long lifespan. Chilled and extended semen is similar except sperm motility and longevity are diminished by chilling and shipping or storage time. Frozen thawed semen has limited number of live sperm, has greatly reduced motility and sperm have a lifespan of only about a day after thawing.

Semen from an infected male can be a source of infection for the bitch. Infection may be suspected or confirmed by a medical history, physical examination, and testing of the male. Infection should be considered if semen has unusual numbers of red and white blood cells with or without bacteria.

Compared with semen from many other species, canine semen is not degraded by short term exposure to room temperature. Nevertheless, it should not be left at room temperature for long periods. Semen is ejaculated in several fractions some of which contain sperm and some of which do not. The sperm rich fraction should be separated from other fractions, and dilution of extended chilled semen or frozen thawed semen with prostatic fluid is not desirable.

If semen is collected from more than one male or if it is stored or shipped to another location, semen containers must be always be marked with positive identification of the male from which it was collected so no misuse will occur.

Techniques for AI in the canine place sperm in the forward part of the vagina or in the uterus. The vaginal environment is not hospitable to sperm. Sperm which are left there survive for only a few hours. In contrast, the inside of the uterus is supportive. So a goal of insemination is to get a large number of motile sperm inside the uterus.

During natural breeding semen is ejaculated into the forward part of the vagina. As mating begins the penis becomes engorged sealing the vagina so semen cannot escape. Ejaculation of a sperm rich fluid fraction is followed by ejaculation of a greater volume of prostatic fluid. Pressure from the continuing ejaculation forces some semen through the cervical canal and into the uterus. Sperm motility also causes movement from the vagina and into the uterus.

Vaginal AI with a standard plastic insemination pipette is a decades old method for insemination of the bitch. This process deposits semen in the forward part of the vaginal space. After insemination, the bitch's rear quarters are elevated so that gravity can assist in keeping the

semen forward and near the cervix. Even though fluid pressure is not great some of the millions of motile sperm will find their way through the cervical canal and into the uterus.

A recent development for vaginal AI is the Mavic ® vaginal insemination catheter Minitube, Verona, WI). This device simulates an inside tie and provides a way to create and hold pressure to enhance movement of semen through the cervical canal. The catheter is currently available in three sizes for use in most breeds. A long, soft plastic outer catheter contains a small diameter flexible metal tube. The plastic catheter has an inflatable bulb at its forward end. After placement into the vagina the bulb is inflated and forms a temporary seal between the catheter and the wall of the vagina. Semen is inseminated through the internal metal tube and reflux is prevented by a check valve. With proper size, placement and inflation, the bitch appears quite comfortable during breeding. Following insemination the catheter is secured with wraps and left in place. After 15 to 30 minutes the wraps are taken off, the rubber bulb deflated and the catheter removed. With good quality fresh or chilled semen the preferred method for AI is vaginal insemination with a Mavic ® catheter or similar device. When semen is poor quality or if it has been frozen and thawed, semen must be placed directly into the uterus by vaginal transcervical catheterization or by surgical intrauterine insemination.

Transcervical insemination (TCI) is the placement of a catheter from the vagina through the cervix and into the uterus. It is best done with a small flexible catheter under direct endoscopic visualization. A non-visual method using a rigid insemination catheter is not recommended. Because TCI is well tolerated by the bitch and does not require sedation, anesthesia, or surgery it is the method of choice for intrauterine insemination. The bitch is gently restrained in a standing position on a table while an endoscope is advanced vaginally so that the cervix comes into view. The end of the scope is carefully manipulated and a flexible plastic catheter is advanced into the opening, through the cervical canal and into the uterus. Semen is inseminated through the catheter into the uterus. Transcervical insemination is a challenging procedure, and placement of a catheter is not always possible. In that case the alternative is insemination by surgical means.

For the veterinary surgeon surgical insemination is a procedure somewhat similar to ovariohysterectomy surgery. Of course, rather than removing the ovaries and uterus the surgeon places a small catheter thorough the wall of the uterus and semen is placed inside. Regardless whether insemination is by TCI or by surgical insemination the semen is placed inside the uterus so conception rates should be identical between the two procedures. Recovery from surgical insemination is very rapid because other than a minor incision in the skin and abdominal wall no other tissues are disrupted. Surgical insemination can be done only once during a single heat so ovulation timing is very important. Transcervical insemination can be done more than once during a single heat, but cervical swelling following catheterization can make multiple inseminations difficult.