

## **Synchronization of estrus of whitetail deer and other select cervids, sheep & goats**

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### **Abstract**

Estrus synchronization in small ruminants is achieved by controlling the luteal phase of the estrous cycle during the breeding season or by inducing premature heat cycles during the anovulatory season. Anestrous animals can be induced into cycling with hormonal treatment or manipulation of the environment. The most commonly used protocol for synchronization of heat in small ruminants involves the use of controlled internal drug release (CIDR) devices, intravaginal sponge impregnated with progestagens, equine chorionic gonadotropin (eCG), and prostaglandin. Prostaglandin based protocols are used during the breeding season. The sudden introduction of a male can also be used to induce and synchronize fertile heat in sheep and goats. Estrus induction and synchronization in deer use similar techniques. This article reviews some successful protocols used to induce heat, synchronize estrus, induce ovulation, and enhance ovulation in small ruminants and deer.

**Keywords:** Estrus, synchronization, reproduction, small ruminants, cervids

The recent increase in interest in small ruminants and the high value of deer has raised the demand for veterinary reproductive services for those species. The development of synchronization protocols has allowed other advanced reproductive techniques such as artificial insemination (AI) and embryo transfer to flourish making valuable genetic material more easily available. Producers are requesting reproductive services that involve intensive reproductive management including synchronization of estrus, off season breeding, AI and embryo transfer. This paper will review available tools that can be used to improve reproductive efficiency in small ruminants and deer.

Estrus induction protocols have the objective of synchronizing heat, advancing the breeding season and both induce and maintain off season cyclic activity. Estrus synchronization techniques in small ruminants must not only establish a tight synchrony, but also provide an acceptable level of fertility upon AI or natural mating during the natural mating season and also the non-breeding season.

Small ruminants and cervids are seasonal polyestrous breeders with reproductive activity centered during the fall and winter. The reproduction of these species is controlled by the variations in daylight (long vs. short days). The effect of light on reproduction is influenced by the number of hours of light and by the time of exposure. Short day breeders can be induced into cycling by reducing the exposure to light (confinement inside a dark building) or by the administration of melatonin implants.<sup>1</sup> Progesterone implants can also be used for induction of fertile heat out of the breeding season<sup>2,3</sup> by mimicking the function of the corpus luteum (CL).

### **The male effect**

The sudden introduction of a male or androgen-treated castrated male can induce and synchronize heat in anestrous ewes and does.<sup>4,5</sup> This is known as the “male effect”. This synchronization is less predictable than that achieved by hormone treatment but is economical and fertility rates are high. The effects are mediated through changes in pulsatile gonadotropin releasing hormone (GnRH) release followed by an increase in luteinizing hormone (LH). This will cause ovulation (silent heat) in two to four days that is characterized by low fertility and premature regression of the CL. The second ovulation (five to seven days later) is of normal fertility and followed by a normal luteal phase. Olfactory response may be a leading stimulus for estrus induction but other factors like tactile, auditory and visual stimuli are also involved. In sheep, preventing the vomeronasal organ from functioning does not affect the female responses to male odor suggesting that the accessory olfactory system does not play a major role in estrus induction. Female responses also seem to depend on previous experience. The responsible pheromone is present in wool and buck hair clippings but not in urine and it is not associated with the buck odor during the breeding season.<sup>6</sup>

The male effect can be used successfully in estrus synchronization programs that use exogenous progestins with or without the use of eCG. The introduction of a male in association with estrus induction protocols using progesterone will increase the number of females bred/marked. The addition of exogenous progesterone at the time of male introduction can reduce significantly the number of short estrous cycles and extended the period from male induction to ovulation in sheep increasing ovulation rate in goats.<sup>4</sup> The uterus is also implicated in the premature regression of the CL after the male effect as both hysterectomy and inhibitors of prostaglandin F<sub>2α</sub> suppressed short cycles.<sup>4</sup>

### **Estrus synchronization during the breeding season**

Estrus synchronization is easily accomplished during the breeding season. The opportunity for control is greater during the luteal phase, which is longer and more responsive to manipulation than the follicular phase. Synchronization protocols during the natural breeding season are based on either the prolongation or shortening of the progesterone phase of the cycle. Prolongation of the cycle is usually accomplished by supplying exogenous progesterone with the use of vaginal implants or oral progesterone. Alternatively, the cycle can be cut short prematurely by regressing existing CLs with the use of prostaglandin.<sup>7</sup>

#### **Use of prostaglandin for estrus synchronization**

A CL must be present in the ovary for prostaglandin-based protocols to work. The control of the estrous cycle with prostaglandin is achieved by termination of the luteal phase through regression of the CL. Natural prostaglandin and its analogs can be used alone or in combination with progesterone or male-effect for estrus synchronization during the breeding season. A typical protocol for synchronization with prostaglandin is to give two injections of prostaglandin nine days apart for sheep and eleven days apart for goats. The typical dose for dinoprost tromethamine is 10-15 mg IM and for cloprostenol the commonly used dose is 50-125 mcg IM. Ewes should be bred 10-12 hours after being marked by teaser ram (around 42-54 hours after last injection).<sup>8</sup> Does should be in heat and ready to be inseminated at 48-52 hours after the last treatment with prostaglandin.<sup>9,10</sup> Prostaglandins are not expensive and provide an economical method of shortening the breeding season in natural mating situations.

The addition of progesterone and eCG (250-500 IU IM) to prostaglandin treatments shortens the interval to heat when compared with females not receiving these supplemental drugs. Breeding should be adjusted to this variation and the use of teaser males is strongly recommended.

The Ovsynch protocol, similar to the program used in cattle, has proven to be successful in inducing synchronized fertile heat in does. It allows fixed-time insemination in goats during the breeding season. The typical protocol involves the injection of GnRH (50-100 mcg IM; considered day zero) followed by a luteolytic dose of prostaglandin (e.g. 125 mcg of cloprostenol IM) on day seven and another dose of GnRH on day nine. Males may be introduced and breeding allowed take place on day eight. Breeding by AI is performed 16 hours after the second GnRH dose.<sup>11</sup>

Estrus induction with progesterone impregnated sponges or CIDRs is done by placing the progesterone carrying device in the cranial vagina of the doe for 12 days. On day 10 the animals are injected with a luteolytic dose of prostaglandin and receive 250-500 IU of eCG. Artificial insemination is performed 42-46 hours after sponge removal. In sheep the sponge or CIDR is left in place for 14 days and eCG is given at progesterone removal. Animals should be exposed to fertile males after progesterone withdrawal.<sup>10</sup>

### **Estrus induction and synchronization outside the breeding season**

Several methods have been used to induce and synchronize heat during the off season. The male effect, artificial lighting (to stimulate decreasing day length) and melatonin are successful in inducing heat in anestrus females. These techniques do not provide tight synchrony, but they permit out of season breeding, are readily available and economical. Prostaglandin-based treatments do not work in non-cycling animals (off season) and are not recommended. The most effective treatments are those using progesterone releasing devices in conjunction with gonadotropin. The use of the male effect in

conjunction with these protocols will affect the time to ovulation and may increase the ovulation rate in goats. This effect needs to be taken into consideration if breeding is done by timed AI.<sup>4,12</sup>

Melatonin implants mimic short days and are used to induce out of season cycles. They are commercialized in Europe under the trade names of Regulin® and Melovine®. They are used after at least two months of increasing daylight and will advance the breeding season by 30-45 days. It increases the ovulation rate and prolificacy by as much as 25%. Melatonin treatment can be used at any time during the anestrous season in breeds less influenced by season.

During seasonal anestrous the average ewe/doe is not cycling and serum progesterone is less than 1 ng/ml with negligible ovarian activity. Progesterone is the natural hormone that sustains pregnancy and is produced by the CL in the ovary following ovulation. Artificially introduced progesterone will mimic pregnancy and will keep the animal in artificial anestrous, preventing ovulation and keeping the female out of heat. When the progesterone source is suddenly removed, the animal will ovulate within a very predictable period even during seasonal anestrous. The effectiveness of a progesterone to serve as a synchronization agent is related to its ability to delay estrus and ovulation and is influenced by the route of administration, duration of treatment and the speed of withdrawal. Alternatively, hormones may be used in various combinations and dosages to induce follicle development and ovulation.

A typical protocol involves the use of a progesterone releasing device (e.g., vaginal sponge impregnated with 45 mg of fluorogestone acetate) that is left in place for 11 days and eCG (250-500 IU) 48 hours prior to sponge removal. Insemination with frozen-thawed semen should be performed 24 h after the onset of estrus.<sup>13</sup>

Several studies have been conducted to evaluate the usefulness of a combination of human chorionic gonadotropin (hCG) and eCG (PG 600, Intervet/Schering-Plough Animal Health, Summit, NJ) as a source of eCG. PG 600 is the only veterinary-grade source of eCG readily available in the US and is marketed to induce cycling in prepubertal gilts. The product is a mixture of chorionic gonadotropins with 200 IU of hCG and 400 IU of eCG/5 ml. The typical extra-label use is based on the amount of eCG that is desired.<sup>14-17</sup> Lambing rates of 36 to 59% have been reported when PG 600 was used to replace eCG. Time to estrus and ovulation in ewes treated with PG 600 in conjunction with intravaginal sponge is 47 h (24-60 h) and 83 h (60-112 h), respectively<sup>15</sup>. The typical dose is 400 IU of eCG. Superovulation has been reported when 1200 IU of eCG was used. Both eCG and PG 600 have the potential to induce the formation of ovarian cysts.<sup>18</sup>

The oral progestagen melengestrol acetate (MGA) has also been used to synchronize heat and induce cyclicity outside the natural breeding season. Melengestrol acetate can be used in combination with gonadotropins, male effect and prostaglandin (during the breeding season) and is fed in doses of 0.125 to 0.3mg/ewe/d for at least seven days prior to introduction of fertile rams. Treatment with MGA increases the percentage of ewes conceiving early in the breeding period and lambing rates.<sup>3,17</sup>

### **Estrus synchronization in deer**

It is unwise to try to group all cervids for the purpose of estrus synchronization. There is no “typical” deer which we could use as an example for the purpose of our discussion in this paper. We strongly encourage the reader to search specific information for the species and subspecies that you will be working with. However, some similarities will be used to describe the procedures being used currently to induce and synchronize estrus in deer. Cervids are represented by over 40 species with 200 subspecies. They are characterized by a huge diversity in morphology, physiology, ecology and geographic distribution. Most cervid species give birth at the time of the year that provides the best chance for the survival and growth of offspring. This adaptation to the natural environment has exerted considerable influence on their reproductive physiology. Species adapted to the northern temperate or arctic regions typically conceive in autumn and calve in the following summer. Species located in the tropics often exhibit limited seasonality or are completely aseasonal. It is accepted that seasonality is affected by endogenous recognition of photoperiodic changes with the majority of species cycling during decreasing day length (short day breeders).<sup>19</sup>

For best results and survival of the offspring breeding should follow the natural cycle. The doe's first ovulation of the year is characterized by silent ovulations and short-lived (8-10 days) CL. A true first cycle follows this first ovulation. Males should be placed with females the as they will continue to cycle until pregnant. Farmers may require buck removal to avoid late breeding in the season. These decisions are made based on management and financial considerations and not on the physiological potential of the specie.

Estrus in the white-tailed doe lasts approximately 24 hours and, if not bred, she will cycle again in 21-29 days. Female deer do not show overt signs of receptivity when in heat in the absence of a male, challenging the producer's task of keeping accurate reproductive records. Controlled breeding is possible with the best time to breed being between 18 and 36 hours after the beginning of a standing heat. Due to these challenges, the best chance to have successful breeding with AI is to breed by appointment.

In many ways, the cervid estrous cycle is similar to that of domestic ruminant species. Thus, artificial estrus synchronization is done in a similar way as it is performed in small ruminants and cattle. Drugs used for manipulation of the reproductive cycle in small ruminants and cattle can be used in a similar manner in deer. Heat synchronization is done using prostaglandin, GnRH, gonadotropins, MGA, vaginal sponges, CIDRs, or ear implants as described above.<sup>20</sup>

Prostaglandin is effective if used during the breeding season when CLs are present and active in the ovaries. Cloprostenol is used at 125-250 mcg/doe IM at CIDR removal after 8-12 days or it is given as two injections 11 days apart.<sup>21</sup>

Timed AI should be done following proven protocols. The most reliable protocols rely on the use of an exogenous source of progesterone in the form of a CIDR or progesterone impregnated sponge. The amount of progesterone and the size of the CIDR are chosen based on the species and size of the doe. For larger species the CIDR-b (1.9 g progesterone) is utilized, whereas the CIDR-g (0.33 g progesterone) is used for smaller deer. The time of exposure to the progesterone delivering device also varies depending on the species. The device is left in place between 8 and 14 days and then removed manually.<sup>21-25</sup>

Equine chorionic gonadotropin is often used at CIDR removal with the final dose depending on the species. It provides for better synchronization of estrus and ovulation (see Table). Higher doses may result in superovulation and excessive number of fetuses.

## Conclusion

The ultimate aim of any estrus synchronization method is to allow AI at a predetermined time after the end of treatment. This requires a very tight synchronization of estrus which can be achieved after careful protocol selection and taking into consideration herd composition, location, level of management, health and nutrition of each individual farm operation.

Table: Recommendations for estrus synchronization in various deer species<sup>22</sup>

Species	Progestin Device	CIDR Duration	eCG Dose (I.U.)	Time for AI after CIDR Removal
Wapiti	CIDR-b	12–14 days	190–200	60–66 hr
Red deer	1 or 2 CIDR-g	10–12 days	150–200	56–60 hr
Sika deer	CIDR-g	12–14 days	50	58–62 hr
Fallow deer	CIDR-g	12–14 days	0	63–67 hr
White-tailed deer	CIDR-g	12–14 days	100–150	60–65 hr

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