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## **Purpose of the Intracytoplasmic Sperm Injection Program**

The intracytoplasmic sperm injection (ICSI) program is offered as a means of establishing pregnancies from oocytes (eggs) recovered from donor mares. Using ICSI, oocytes are injected with individual sperm from a donor stallion, and the resulting embryos are allowed to develop in the laboratory for approximately one week. Developed embryos are transferred into synchronized recipient mares.

This advanced reproductive technology is appropriate for mares that are unable to become pregnant themselves (i.e., mares with chronic uterine disease, cervical lacerations, or other damage to the reproductive tract that prohibit the mare from conceiving or supporting an embryo in the uterus for any length of time). The procedure can also be attempted when pregnancies are sought from limited sperm supplies.

The procedure should only be used on mares that are not suitable candidates for routine embryo transfer (i.e., mares in which viable embryos are seldom recovered from standard uterine flushing), or, if done to obtain foals from a given stallion, for sperm that cannot be utilized effectively with standard insemination techniques.

Because of the expense of the technology involved and the amount of labor associated with ICSI, foals produced from this program should be valuable enough to justify the increased effort and expense to produce offspring. Before participating in the ICSI program, it is important for each owner/lessee to know the regulations of their breed registry regarding the possibility of registering any resulting foals.

## **Overview of the procedure**

Intracytoplasmic sperm injection is a recently developed technique in which oocytes (eggs) of a valuable broodmare (donor mare) are fertilized in the laboratory. The oocytes are recovered from the mare's ovarian follicles, and then cultured to induce maturation, mimicking the developmental changes that would occur in an oocyte during the day or so immediately before ovulation. This maturation process generally takes 12 to 30 hours, depending upon the stage of maturation of the recovered oocytes.

Matured oocytes are injected with individual sperm from the desired stallion.

For this procedure, one sperm is injected into the cytoplasm of each oocyte under a high-power microscope. The resulting fertilized oocytes are cultured in the laboratory for 7 to 10 days, to allow development into blastocysts, that is, embryos suitable for transfer to a recipient mare.

This contract with INFOAL, INC includes recovery of oocytes, fertilization, embryo culture and transfer of resulting embryos.

## **Anticipated results**

When transvaginal aspiration of all follicles is performed, we typically recover oocytes from 60 to 70% of the follicles aspirated; in fertile mares, this averages 6 to 8 oocytes per aspiration session. About 60% of these oocytes may mature in the laboratory and be injected with sperm (average of 4 per session). We anticipate a 20% rate of blastocyst development if the sperm is from a fertile stallion, thus approximately 80% chance of a transferrable embryo per aspiration session.

These anticipated rates may decrease markedly with:

. Mare age: Old mares have fewer follicles, and oocyte recovery rates are lower.

. Infertile mares: Some causes of infertility appear to be related to poor oocyte quality.

. Subfertile stallions: The embryo development rate after ICSI is lower with some stallions.

When oocytes are recovered from the one preovulatory follicle, a 75% recovery rate is expected; however, only one (or occasionally two) such follicles develop per cycle.

## Benefits of the program

When multiple oocytes are recovered, all oocytes that form embryos may be transferred to separate recipient mares, and thus have the potential to produce a foal. In addition, the ICSI program can utilize equally frozen, fresh, or cooled, transported semen, and sperm of low numbers or low quality.

## The following information is important:

a. How many ICSI pregnancies do you want from this donor mare?

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b. Name, address, and phone number of primary contact regarding this mare.

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c. Name, address, and phone number of contact for collection and shipment of semen, and name of stallion:

Stallion 1:

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Stallion 2:

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