IMPACT OF HISTEROSCOPY IN THE MARE IN DAYLY PRACTICE

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Most reproductive abnormalities in mares can be detected using common procedures such as palpation, vaginoscopy and ultrasonography and are sometimes supplemented by microbiological culture, cytology and biopsy. However, sometimes it is necessary to apply more sensitive procedures to achieve a definitive diagnosis and/or to objectively classify the severity of a problem when interpreted with ultrasonography. One of these techniques is endoscopy, which provides a real-time visualization of the inside of the uterus without the need for interpretation[1,2,3].

Endoscopy is commonly used in equine medicine for observation of the upper respiratory system and the first portion of the digestive system such as oral cavity, esophagus and stomach. In the mare an endoscope can be used to detect intrauterine abnormalities that most of the time are not observable by ultrasound such as adhesions, endometrial cysts, foreign bodies, volume increases, hemorrhages, biofilms, mucus, retention of endometrial cups, etc. Also an endoscope achieves images with more detail of problems at the vaginal level such as vaginal varicose veins, lacerations and accumulation of urine in the vaginal fundus as well as a more accurate evaluation of cervical integrity. In addition, an endoscope can be used to perform deep artificial insemination with low doses of semen on the utero-tubal papilla[1,2].

It is an uncommon procedure and therefore not routine in the daily practice of the reproductive clinic in mare. Since the necessary endoscopy or video-endoscopy equipment demands a high investment, often difficult to transport to the field and need adequate facilities for a good examination and proceed[1].

In 2017 in Bioteq®, we acquired a video-endoscope and in these 6 years we have performed a total of 147 hysteroscopies in mares of different breeds and ages. 130 of them (89%) were performed for a diagnostic purpose in mares with a history of subfertility or infertility and 17 of them (11%) for a therapeutic purpose in mares with an undergoing problem and already diagnose. Of the 130 diagnostic hysteroscopies, 82 of them did not have a previous ultrasound finding, so 56% of the cases had a finding only with endoscopy. Of the 44% (65 endoscopies) who had a previous ultrasound finding, in 37% (24 cases) their previous ultrasound finding was of lower clinical magnitude in its interpretation than the observed reality and in 63% of cases (41 cases) endoscopy revealed and confirmed something very similar to what was observed by ultrasound.

This denotes and confirms the importance that this real-time image generator tool can have and the potential it offers to make correct therapeutic decisions and therefore better prognoses.

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Nowadays he has founded a new and innovative project concerning sex sorting semen using nano-technology and creating his new business BioseXYng®.

Recent publications-presentations

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Biofilm: is it real and how do we manage it in equine reproduction

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Bacteria utilize numerous methods to survive degradation by the host immune system and antibiotic therapy. One survival tool utilized by bacteria is the production of a biofilm. Biofilms allow bacteria to be unrecognized by the host immune system, prevent exposure to antibiotics, and allow for exchange of genetic material leading to antibiotic resistance. Until recently very little was known about the importance of biofilm in cases of equine endometritis. Recent studies have identified bacteria residing in a biofilm on the endometrial surface of mares using a model of endometritis. Currently there is not a good diagnostic technique for detecting a biofilm associated infection in clinical practice. Clinicians should strongly suspect a biofilm in mares that are identified to have an infection, appropriate treatment is applied to the case and the infection is not readily cleared.

Biofilm infections have a high rate of failure due to the bacteria being protected from antibiotics by being in the biofilm lifestyle. Non-antibiotics can be effectively used to disrupt the biofilm that is protecting the bacteria allowing antibiotic treatments to effectively eradicate the infection. Caution should be used when mixing antibiotics with non-antibiotics as many were found to result in inactivation of the antibiotics used in the treatment. Commonly used effective compounds are presented in Table 1.

Overall the incidence of biofilm or latent bacteria is unknown in the broodmare population. However, with overall pregnancy rates in the broodmare population being relatively high per cycle it is suspected that the incidence rate of biofilm or latent bacteria is low. In individual mares failing to become pregnant biofilm and latent bacteria should be a consideration as a cause of subfertility. Fortunately, research has described how these infections might develop, improved diagnostic techniques and effective treatment strategies for biofilm or latent bacterial endometritis.

Table 1. Antibiotic and Non-Antibiotic Combinations For The Treatment Of Biofilm Associated Bacterial Endometritis In Mares

Tris EDTA- final concentration in the syringe should be 50 mM Tris and 3.5 mM EDTA Note: Tris-EDTA and Tricide are similar; however Tricide is not equivalent to Tris-EDTA in regards to bacterial killing To make Tris-EDTA: 16oz bottle of Dechra Triz-EDTA crystals; add 8 oz of sterile water (this is different than the bottle instructions). The 2x concentration of Tris-EDTA solution will be further diluted by the antibiotics below to the proper final concentration.					
Antibiotic	Drug Amount	Tris EDTA	QS	Final volume	Notes:
Amikacin (250 mg/ml)	4 mls (1 gram)	30 mls	16 mls sterile fluid (Saline, LRS, Sterile H_2O)	60 mls	10 mls of 8.4% sodium bicarbonate should be added to the amikacin
Ceftiofur (1 gram reconstituted in 20 mls)	20 mls (1 gram)	30 mls	10 mls sterile fluid (Sterile H_2O)	60 mls	

Ciprofloxacin (10 mg/ ml)	40 mls (400 mg)	40 mls	0	80 mls	Split between two syrin-ges
H_2O_2- 1% final concentration in the syringe A 3% stock solution is available at many drug stores and veterinary distributors					
Antibiotic	Drug Amount	H_2O_2	QS	Final volume	Notes:
Amikacin (250 mg/ml)	4 mls (1 gram)	20 mls	26 mls sterile fluid (Saline, LRS, Sterile H_2O)	60 mls	10 mls of 8.4% sodium bicarbonate should be added to the amikacin
Ciprofloxacin (10 mg/ ml)	40 mls (400 mg)	20 mls	0	60 mls	
DMSO- 30% final concentration in the syringe 99% stock solution is used for calculations below					
Antibiotic	Drug Amount	DMSO	QS	Final volume	Notes:
Ceftiofur (1 gram re-constituted in 20 mls)	20 mls (1 gram)	20 mls	20 mls sterile fluid (Sterile H_2O)	60 mls	
Ciprofloxacin (10 mg/ ml)	40 mls (400 mg)	20 mls	0	60 mls	

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COMMON REPRODUCTIVE PROBLEMS IN STALLIONS

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Similar with the literature on our clinical experience problems related with spermatogenesis (testicle degeneration) and ejaculatory dysfunctions are the most frequent reproductive problems of stallions. Most of problems are related with disturbs on testicle thermoregulation. Equine Piroplasmosis (Babesiose) is endemic in most states from Brazil and is an important factor related with the testicle degeneration installation because of high fever induced by the disease. The increase on testicle temperature will induce an increase on cell metabolism with a consequent increase on oxygen needs. As the blood supply to testicle is poor and few oxygen is available the cell dies. The sequence of sperm abnormalities is related with the severity of the injury. The evolution of the sperm abnormalities and daily sperm production is important to determine a testicle degeneration prognosis that usually is favorable. The hormonal profile can also help to determine the degree of testicular damage being important to measure the estrogen levels. The recovery of normal testis tone and sperm production is possible in most of cases if the cause is discharged. The use of anabolic steroids is also related with disturbs of semen production causing a more profound reduction in scrotal circumference. Disturbs of sexual behavior are the second more frequent clinical problems observed in our ambulatory routine of horse andrology and most of them are characterized by ejaculation dysfunction, these problems can have a physical or psychogenic origin. Most of times is difficult to make a differential diagnosis between physical and psychogenic problems. On psychogenic cases the adoption of good practices of reproductive management are important for the recovery. On cases where the problem is physical is important to determine if pain is interfering with the sexual copulatory function, and several strategies (pharmacological and management) can be adopted to help the stallion to achieve ejaculation. Other observed problems are pathologies of the internal glands as vesiculitis and blocked ampullas. The treatment of vesiculitis is most of time frustranting with a high probability of recurrence. The ejaculated most of times contain large amount of blood and inflammatory cells. For blocked ampullas the differential diagnosis with testicle azoospermia is made with the dosage of alkaline phosphatase levels on the seminal plasma. The treatment involves sequential semen collection associated with trnsrectal massage of the ampullas. The principal penile pathology is the penile haematoma induced by trauma during the breeding or semen collection on this case its very important to drain the blood, hydrotherapy and the penis must remain suspended.

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HOW TO PROCESS SEMEN FROM DIFFICULT STALLIONS

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Selection of stallions is performed by phenotypic assessments, such as the conformation of the animal and their athletic performance,[9] unlike bovines where reproductive parameters are assessed in bulls before becoming a commercial sire. Another factor that affects sperm quality is age, and often owners decide to freeze semen from stallions that have advanced age and may be subfertile. Also is knowed that some breeds as Spanish and Mangalarga stallions are less resistant to semen preservation.

Consequently, several techniques have been developed to increase the sperm quality of stallions. Stallions whose semen does not resist the centrifugation process, the use of such techniques as SpermFilter, and cushioned centrifugation may be beneficial. For stallions whose semen has a low resistance to cryopreservation, the use of extenders with specific cryoprotectants, such as amines, in general improve sperm cryosurvival and fertility.

When the initial quality of semen is poor, sperm selection using commercially available density gradients, such as EquiPure (Nidacon) or Androcoll-E (Minitube) can be performed before cryopreservation.These gradients select the sperm that exhibit better progressive motility, cell integrity, and no morphologic defects . Another alternative is to select sperm after thawing. For this procedure, the contents of four straws are gently layered on top of 2 mL of Equipure, centrifuged at 300 to 400 G for 20 minutes, and the pellet resuspended with the freezing extender. Based in our experience this protocol improves the motility and the fertility of frozen semen of some stallions. It is postulated that the removal of bad quality or dead sperm that can generate oxidative reagents can improve the fertility.

Cushion centrifugation (CC) is an alternative technique for removing SP from semen. This method aims to maximize the sperm recovery using a high G force (1000 G/ 30 minuts). Another possibility for removal of seminal plasma is to use a filtering method composed of a synthetic hydrophilic membrane (Spermfilter , Botupharma Brasil)) that retains the sperm cells and allows for the passage of SP , concentrating the sperm cells with less damage and also less cell compactation. Recently publications have been shown an increase on fertility of cooled semen that was reprocessed by Spermfilter after 24 hours of refrigeration.

Methylformamide has been used as the preferential cryoprotector for Stallion semen it causes less osmotic damage to sperm than glycerol because of the lower molecular weight and viscosity.[7] For stallions with semen that has satisfactory freezability ("good freezers"), the use of extenders containing dimethylformamide or methylformamide may not result in a significant increase in post thaw sperm motility; however, it does increase the fertility of the frozen semen. In stallions whose semen has low resistance to cryopreservation ("bad freezers"), the use of extenders containing dimethylformamide or methylformamide provides a significant improvement in sperm motility and fertility compared with extenders containing only glycerol. The use of a combination of cryoprotectants affords better protection to sperm compared with the use of single agents. Several equine reproduction centers in Europe, the United States, and Brazil have preferentially used a commercial extender (BotuCrio), which includes a combination of methylformamide and glycerol.

For semen cooling and transportation, the use of the new formulation of extenders containing casein and cholesterol have been show to increase the cooling ability and also membrane integrity. Casein is a milk protein that avoid the binding of seminal plasma proteins that usually destabilize the sperm membrane inducing damage and precocious capacitation. Cholesterol increases membrane fluidity improving the ability of the sperm cell to resist to osmotic stress.

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What's new for embryo transfer?

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The collection and transfer of horse embryos (ET) was first reported in 1972 [1-2]. The uptake in equine clinical practice was relatively slow, until an upsurge in ET in Argentina during the 1990s, primarily because of the increasing commercial demand for high-goal Polo Pony horses, combined with a shift from non-surgical to surgical transfer [3-4]. Since then, ET has become the most popular assisted reproductive technique (ART) in the horse. During the current decade, the ET industry is shifting towards the use of in vitro produced embryos, especially in sport horse breeds, owing to the increase in the efficiency of OPU-ICSI: an ART capable of producing several embryos per session with the use of a very small amount of sperm. Furthermore, this technique allows the successful cryopreservation of embryos which results in a more efficient management of recipient mares and marketing of embryos worldwide [5]. The most relevant factors involved in the success of this technique are: 1) the number of recovered oocytes per session, 2) the donor mare (individual variation) and 3) the ICSI lab's experience [5]. Regarding the production and collection of in vivo derived embryos, the quality of sperm, age of donor's mare, and the operator's experience and flushing technique [6] appear to be the main factors that influence embryo recovery rates. Furthermore, several studies have shown a beneficial effect of the length of estrus on both embryo recovery (donor mares) [7] and post-transfer pregnancy rates (recipient mares) [8]. The correlation between length of estrus and fertility appears to be related to the development of a more receptive endometrial environment for embryo survival. Advances in the cryopreservation techniques of in vivo derived blastocysts seem to have overcome the dogma about unsuccessful pregnancy results following cryopreservation of embryos >300 μm. These advances are: 1) the collapse and aspiration of blastocysts by micromanipulation prior to cryopreservation [9]; and 2) the use of a new vitrification technique that allows successful cryopreservation of blastocysts of up to 500 μm without the need of previous collapse [10]. Finally, the development and refinement of recombinant equine FSH (reFSH) has produced an apparently practical and consistent solution to the lifelong quest for super-ovulation in donor mares [11]: Treatment of deep anestrous mares with 1.3 mg of reFSH once daily for 7 days resulted in the mean ovulation and recovery of 5.5 follicles and 2.6 embryos per mare, respectively. However, this product is yet to become commercially available. The selection of suitable recipient mares and the technique of ET are key points to the success of a commercial embryo transfer program. While the transfer of in vivo derived embryos allows a wider window of donor-recipient asynchrony without compromising pregnancy rates (recipients may ovulate between 1 day before and up to 4 days after the donor mare without differences in pregnancy rates), the optimal window for in vitro produced embryos is much shorter (i.e. 24 to 36 h), obtaining best results from transfers to Day 4 recipients [12]. A recent study, contrary to previous belief, has shown that the vascularization of the CL of the recipient mare, as determined by doppler ultrasonography, at the time of transfer is positively correlated with pregnancy [13]. Finally, the transfer of embryos with the aid of a speculum and cervical forceps, as described by Wilsher and Allen in 2004, has recently shown to consistently improve post-transfer pregnancy rates and reduce variation in pregnancy results amongst operators, compared with the traditional manual technique [14].

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Ovulation induction and failure in mares: What's new?

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Human chorionic gonadotropin (hCG) is still the most used ovulation induction drug in the mare, at least in Europe. Doses of 750 to 3000 IU (administered subcutaneously, intramuscularly, and intravenously) have been proven to be successful at inducing ovulation within 48 h of treatment. However, it is well known that its efficacy decreases after the second treatment when administered in the same mare and season, due to the development of antibodies against hCG. In some mares, these antibodies may last for several years. Therefore, some studies have reported a reduction in the ovulatory response to hCG in aged mares (i.e. > 10 years old). As a result, the use of GnRH agonists has become popular as an alternative to hCG to induce ovulation in the mare. Several compounds of different potency, formulations and doses have been tried successfully in the mare for induction of ovulation. Deslorelin, buserelin, and histrelin have been used successfully as single treatment and are commercially available (either licenced for horses or humans) to induce ovulation in mares. Most recently, tripterolin has been shown to induce ovulation in mares as efficaciously as hCG or buserelin. The efficacy of GnRH agonists in inducing ovulation is reduced in spring transitional mares, as the LH pituitary reserves are low in non-cyclic mares. Therefore, in transitional mares, a combination of hCG and a GnRH agonist may be used to improve timely ovulation induction. In cyclic mares, higher doses of GnRH agonists are often needed when the drug is produced in a slow-release (long-acting) formulation (i.e. >1.25 mg per treatment; for example Ovuplant®: 2.1 mg; Sucromate®: 1.8 mg; BioRelease®> 1.25 mg, etc.). On the other hand, GnRH agonists formulated in saline or aqueous excipients can induce ovulation at a much lower dose. Buserelin (Suprefact®) at a dose as low as 125 µg (0.125 mg) in single treatment induced ovulation within 48 h in 85% of treated mares [1]. Although there have not been direct comparisons between hCG, GnRH agonists in aqueous and in slow-release formulations, it appears that hCG and GnRH agonists in aqueous solutions induce a slightly lower ovulatory response within 48 h when treated in a random population of mares (breeding stud, and varies between 78 and 90%) compared to slow-release formulations of GnRH agonists (deslorelin) which report ovulatory responses within 48 h from 87 to 93%, despite earlier start of treatment (i.e. follicular diameter at induction of ≥30 mm in diameter). However, the latter compounds are usually not available in many countries and their cost is high compared with hCG and other GnRH agonists licenced for humans (in aqueous formulations). Nevertheless, following the use of ovulatory induction drugs in mares, ovulatory failure or delayed ovulation are relatively common and frustrating outcomes. It is important to differentiate amongst these two outcomes, as the mechanisms underlying the causes are completely different. In the first place, and the most likely outcome, is the delayed ovulation. In this case, the ovulation induction drug is administered in what is believed to be the right moment: a growing dominant follicle of 35 mm or larger in diameter, from an estrous mare with obvious endometrial edema. However, the mare does not ovulate within the expected time frame after induction (36 to 48 h). On the contrary, the follicle continues to grow, and the endometrial edema remains similar or increases in intensity for an extra 24 to 96 h, before the follicle finally ovulates. This represents typically a 7 to 20% of induced cycles in cyclic mares, and the main reason for this is that the mare was too far from spontaneous ovulation to respond to the ovulatory drug, despite having a follicle size of what is considered enough to respond (and most mares would) to the ovulatory induction drug. The statistical analyses of large data sets of mares that did not respond to the ovulatory induction drug in a timely manner often show that the single most relevant factor was the follicle diameter at the time of induction (significantly lower diameter compared to that of responding mares). The percentage of delayed ovulations is significantly increased in transitional mares, before the first ovulation of the breeding season, owing to the reduced LH reserve of the pituitary in spring transitional mares. The second possible outcome is a true ovulatory failure, and this has two possible presentations. In the first presentation, the leading follicle on which the drug was administered and intended to induce ovulation, does not ovulate but undergoes atresia or "follicle regression". These follicles are more commonly found in a) spring transition, b) mares with two major follicular waves (diestrous follicles), and less frequently c) in post-partum mares which after producing a dominant sized follicle during the foal heat, enter a temporary anestrous phase in which the follicle remains static or regresses. On such occasions, the regressing follicles do not luteinize despite showing a varying number of echoic specks within the antrum, and the mare feels anestrous on rectal palpation unless it has a CL (option b). The second presentation to a true ovulatory failure, is the development of a hemorrgahic anovulatory follicle (HAF), in which the oocyte is not released, and fertilization is not possible. This may occur in up to 5-8% of all cycles, with an increased incidence during the months of the peak breeding season (June-July in the N.H.). In fact, the ovulatory drugs function well, as the HAF cycles has the similar LH and estradiol hormonal patterns as ovulatory cycles. The HAF luteinizes and functions as a CL, producing progesterone, and therefore the mare enters a standard period of diestrus (13 to 16 days), after which the HAF regresses and the mare comes back in heat and a new dominant follicle can be induced. The aetiology of HAF still remains unknown. However, in an experimental model, they were reverted to ovulatory follicles by intrafollicular administration of PGE_2 and $PGF_{2\alpha}$.

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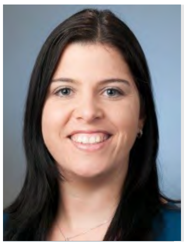
Juan graduated in Veterinary Medicine in Murcia (Spain) in 2003, and did his MSc in Equine Science in the University of Edinburgh (Scotland) in 2006. He completed his Residency in Large Animals Theriogenology in the Royal Veterinary College (University of London, 2008) under the co-supervision of Prof. John R. Newcombe, and became a Diplomate of the European College of Animal Reproduction (Dipl. ECAR, Equine) in 2009. Finalized his PhD in 2010 in the University of Helsinki supervised by Prof. Terttu Katila. Juan worked as Assistant Professor in Equine Reproduction at the Faculty of Veterinary Medicine, Universidad UCH-CEU in Valencia, Spain (2010 to 2016) and in the Veterinary School of Utrecht University, The Netherlands from 2016 to 2019. During his time in academia, he completed several research placements in different laboratories over the world working with Prof. Terttu Katila (Helsinki, Finland), Prof. O.J. Ginther (Wisconsin, USA) and Prof. Luis Losinno and Javier Aguilar (Rio Cuarto, Argentina). After his time in The Netherlands, he worked as stud veterinarian during two breeding seasons in Australia and Sweden. Currently he is working again as Assistant Professor in Facultad de Veterinaria, Universidad CEU Cardenal Herrera, in Spain. His research and clinical interests are breeding management of mares, anovulatory disorders, induction of ovulation, ultrasonography of the corpus luteum, embryo transfer and ovum pick-up in mares, amongst others. He has published over 60 articles on equine reproduction in international indexed JCR journals and lectured in numerous international congresses on equine reproduction over the world.



Real World Approaches to ART in Practice

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Several Artificial Reproductive Technologies (ART) have been used in horses for a few decades, starting with artificial insemination and progressing to embryo transfer, in-vitro produced embryos, and cloning.

Currently artificial insemination is the most common ART used for breeding horses (except for thoroughbreds) and embryo flushing and transfer is the most common way to obtain more than one offspring per mare per season.

During the last 5 years, with the increase in success rates of in vitro fertilization using ICSI (Intracytoplasmic sperm injection) there has been a significant expansion in the use of aspiration of immature oocytes for in vitro embryo production for commercial purposes. This procedure is called TVA (Transvaginal Aspiration) or OPU (ovum pick-up) and recently this practice is getting more and more common, switching from being performed exclusively at specialized centers to becoming available for practitioners in the field.

The TVA of oocytes is performed with mares restrained in stocks, standing and under sedation (usually with a combination of detomidine and butorphanol). The procedure requires the use of a transvaginal ultrasound probe with a needle guide, a tubing system, and a pump. Usually, 2 practitioners will be on the back of the mare; one with the arm in the rectum fixing the ovaries and the other one manipulating the needle to puncture and scrape the follicles. A third person will be infusing the flush media into the follicles. For the procedure to be safe and successful a high level of communication and coordination between all the team members is required. Each follicle is aspirated and flushed several times (6-10) to be able to obtain the oocyte. The procedure is repeated for every follicle visible on ultrasound.

Not every follicle will yield an oocyte. The reported recovery rate of oocytes on TVA varies from 10-30% at the beginning of the learning process to over 50-60% in practices with more experience. The recovery rate can be influenced by individual mare, breed, age and size of the follicles. There is a learning curve for this procedure; practitioners that are new to it start with low recoveries and success significantly increases with practice. Some colleagues suggest that a minimum of 100 aspirations are needed to become successful and confident with TVA.

This procedure is usually very safe but severe complications such as hemorrhage, abscesses, or rectal tears have been reported in a few occasions.

The TVA is more invasive than a traditional embryo transfer and there is an expected level of inflammation in the mares after the procedure, particularly detectable using abdominocentesis that may be subclinical or clinically evident. Clinical signs of pain or discomfort in the first 24hrs are not uncommon.

The number of oocytes obtained per procedure is highly variable but reports show an average 5-13 oocytes per aspiration. There are many factors affecting successful embryo production and one of the most important is the number of oocytes obtained in the aspiration, different reports show an average of 0.6-2 blastocysts per aspiration. The team at Utrecht University reported that one or more blastocyst will be obtained in 78% of the aspirations.

Most embryos are frozen or vitrified without a significant loss in viability, this is an advantage of in-vitro embryo production (IVEP)when compared with in vivo produced embryos.

The success rate for pregnancy has been reported to be between 70-78%, but some of those pregnancies will be lost before foaling (approximately 15%). Based on reported success rates, an in vitro produced embryo will generate a foal 50-60% of the time (Claes and Stout 2020).

An important benefit of in vitro produced embryos is that of time efficiency; TVA/ICSI is a quick procedure and doesn't require the cycle of the mares to be monitored. Additionally, the procedure can be performed year-round and beyond the breeding season.

In vitro production of embryos is becoming the most important way to produce foals out of mares with severe fertility issues (chronic infections, incompetent cervix or infertility of unknown origin) or very busy performance schedule. This technique also provides the opportunity to generate embryos from stallions with very low availability of sperm (low quality or deceased stallions). With the growth trend of these procedures, probably IVEP will be soon the most common way to obtain multiple embryos per year from the same mare.

Ghislaine Dujovne

Dr Ghislaine Dujovne is originally from Chile she graduated from Universidad de Chile in 2004, after graduation she did a year advance training in Animal Reproduction with Equine emphasis at the same institution. After working in private practice for 4 years, mostly in thoroughbred breeding industry she moved to the US to perform a Residency in Equine Reproduction and a Master of Science at Auburn University in Alabama. She obtained her American College of Theriogenologist diplomat status in 2010 and stayed as a lecturer in Auburn University before moving to California. Dr Dujovne works at University of California Davis since 2012 as Associate Professor in Equine Theriogenologist. Her work is mostly clinical in combination with applied research in Mare subfertility and Advanced reproductive technologies. Her daily work is focused in equine reproduction clinical cases at the Veterinary Teaching Hospital, UC Davis; teaching veterinary students and training Theriogenology Residents to become specialists.