Exodus has always attempted to be on the cutting edge of new research as it becomes available, this is one method of keeping our clientele up to speed on the advances in equine reproduction. As more horse people advance their knowledge, the long range benefits become evident with more foals being born, generating more income for all!

For the last 15 to 20 years, it has been promoted that the "cooling rate" is the main ingredient to successfully transporting semen. However, new research has proven that yet the cooling rate is important, the method of how the semen is processed has become more important! *Concentration*, *extension*, *osmolarity** & *pH** are just as important in the overall equation along with the cooling rate. When the sperm concentration is too great, lactic acid* increases at such a fast rate that the pH* begins to lower, which then causes a rapid drop in motility and survivability. Exodus first began to tout the theory of *broad seminal extension*, dating back to 1997 as we began to draft the claims for our first ***Equine Express***® **II** patent. We knew that a slow cooling rate, was only a small factor in the equation. For successful semen transport; concentration, extender, osmolarity & pH all play a major role in the overall success!

All ***Next Generation***® **Semen Extenders** are buffered solutions that are balanced for a specific osmolarity & pH.

Osmolarity:

The concentration of an osmotic solution especially when measured in osmols or milliosmols per liter of solution

***pH:**

A shorthand designation for the concentration of hydrogen ions in a solution, defined as the negative logarithm of the concentration of hydrogen ions. The concentration of pH reflects the total amount of hydrogen ions present in solution.

Alkaline:

A solution whose pH is more than 7.0

Acidic:

A solution whose pH is less than 7.0

***Buffered solutions** and/or **buffers:**

Solutions which resist a change in pH when small amounts of an acid or base are added. However, these two compounds must be able to coexist in a solution, without completely neutralizing each other. A chemical system that prevents changes in Hydrogen ion concentration, **a true buffer is a stable solution of known pH**

***Lactic Acid:**

A hygroscopic organic acid $C_3H_6O_3$ that is known in three optically isomeric forms:

D-lactic acid:

The dextrorotatory form present normally in blood and muscle tissue as a product of the metabolism of glucose and/or glycogen

L-lactic acid:

the levorotatory form obtained by biological fermentation of sucrose

Suggested Off Season A.V. Care

The proper care of an A.V. is only realized when you pull it out of the closet the following spring and realize that it has dry rotted and is no longer any good. We have outlined a simple outline for the proper care of an A.V.

1. Thoroughly clean & rinse with our A.V. Scrubber Gel
2. With the pin out of the valve.
3. Lay A.V. flat in bottom of sink.
4. Pin down towards the drain.
5. Let all excess water drain for 2 - 3 days.
6. Replace pin into the valve, finger tight.
7. Using a small hand/ball air pump.
8. Pump enough air so the sides do not touch: 3 - 4 pumps.
9. Thoroughly cover with baby powder, inside and out.
10. *Place inside of a plastic bag and place in a freezer.

Do not accidentally lay anything on the A.V.

***The current theory of fresh cooled semen transport:

1. 750,000,000 million progressively motile sperm per dose
2. 25 million progressively motile sperm per extended ml
3. Extended to a minimum ratio of 5 to 7 parts semen extender.
4. Intermittently test your stallion for antibiotic sensititivity.
5. Intermittently test the cooling capacity of your stallion's semen.

***Stallions that experience a difficult time with transported semen, usually have an extremely high sperm concentration. Extend these stallions more broadly which should increase the seminal longevity post transport. Be sure to inform the mare owners of your final ratio to keep open lines of communication.

***Antibiotics should be tested on each stallion for sensitivity reactions. This can be easily performed using the Equine Express II cooled semen & genetic preservation transport system and our new Semen Extender Test Kit where you can test your stallion on both of our extenders and our entire line of antibiotic offering. Set up three boxes with six (6) different syringes. For each antibiotic, mark one syringe as your control, mark the other syringe as your variable group. Every stallion has a different level of tolerance to various antibiotics. To maximize your stallion's fertility, know which antibiotic works best for him, by monitoring the progressive motility every (6) six hours.

***When a stallion produces an unusual amount of gel, he's actually telling you that something is not 100% right with your collection procedure. Don't ever be afraid to attempt new collection procedures, whether it be water temperature, pressure or internal lubrication of the AV or height of the phantom. Any of these could optimize your stallion's seminal output and/or quality.

***Keeping a daily record to monitor your step by step collection procedure will benefit you greatly when a change occurs in his semen quality and/or output. A column type notepad will allow you to monitor everything from the AV temperature to number of mares bred on that collection. You can download a copy of this form from our website under Stallion Management Tools. All of this collaborated information will allow you to maximize your stallion's performance and give you a better analysis when a problem does occur which will ultimately allow you the opportunity to prevent a set back in its tracks while, yielding you more paid service fees!

***Nutritional management of your stallion begins at least sixty (60) days before the start of the breeding season. Feeding a well balanced diet, including chelated Copper, Zinc, Manganese as well as Vitamin C, a super source of antioxidants, will/can dramatically improve your stallion's success in the breeding shed. Planning and preparation are the two key ingredients for a successful breeding season, they usually require very little work!

How to prepare a culture plate

Preparing a culture is quite simple once you learn the technique. Having this knowledge can & will improve your conception rate as well as your overall herd management by being able to plate a wound or nasal discharges.

1. After the culture swap has made contact with the area to be cultured and safely back in it's protective sheath.
2. Remove the lid on the culture plate of choice.
3. Pick up base plate with your left hand without allowing your finger tips to extend past the top edge.
4. Carefully remove the swab from it's protective sheath, holding the swab in your right hand.
5. Even though the swab is round, assume that it has four (4) sides.
6. Using a left to right stroke, swab a "Z" pattern on the bottom of the plated media.
7. Rotate the swab counter clockwise after each completed "Z" stroke.
8. After all sides of the swab have made contact with the culture media, discard the swab.
9. Replace the lid on the Petri dish
10. Invert sitting the plate upsides down and set in a incubator set at 37c.
11. By setting the plate upside down, the condensation will accumulate opposite the plated culture media.
12. Checking the progress of growth should be no less than intervals of 6 - 8 hours and the plate should be discarded after 60 hours.
13. If a growth becomes evident where by a series of organisms begin to grow, utilizing the antibiotic disks, apply a series of antibiotic disks using the Antibiotic Application Dispenser - #658
14. We have posted pictures of various bacteria and yeast growths on our website. Make it a point to become accustomed to what common growths look like: www.exodusbreeders.com/bacteria
15. Maximize your reproductive efficiency!

Bacteria Classifications

Gram Positive

Streptococcus: A non-motile chiefly parasitic gram positive bacteria.
B-hemolytic Streptococcus: An aggressive gram-positive bacteria.
Staphylococci: Various non-motile gram positive spherical bacteria.

Gram Negative

E. Coli: A gram negative bacteria commonly found in intestinal flora.
Klebsiella: Plump non-motile encapsulated bacterial rods.
Pseudomonas: A short rod shaped bacteria; fluorescent green pigment.

Plated Media Options

Columbia (CAN) Agar Plate

This plate has Columbia (CAN) Agar, a 5% Sheep Blood as its media of choice, which, covers the spectrum specific to Gram Positive bacteria.

Levine EMB Augar Plate

This plate has strictly Levine EMB Agar as its media which, covers the spectrum specific to Gram Negative bacteria.

MacConkey Agar Plate

This plate has strictly MacConkey Agar as its media which, covers the spectrum specific to Gram Negative bacteria.

Mueller Hinton Plain Plate

A widely used plate in every laboratory, for specific sensitivity testing, Mueller Hinton plates will grow just about any form of bacteria. This is a Multi-Purpose Culture Plate.

Tryptic Soy Auger (TSA) Plate

This plate has Tryptic Soy Agar (TSA), a 5% Sheep Blood as its media of choice, which covers a very broad spectrum of bacterial growth. This is a Multi-Purpose Culture Plate.

Universal Bi-Plate Plates- Columbia CAN & MacConkey Agar

This plate has Columbia CAN & MacConkey Agar as its medias of choice, which, as its name implies, is a universal culture plate that covers the spectrum of both Gram Positive & Gram Negative bacteria. This plate is a Multi-Purpose Culture Plate.

Universal Bi-Plate Plate - Blood Agar TSA 5% & MacConkey Agar

This plate has Blood Agar TSA 5% & MacConkey Agar as its choice of medias and as its name implies, is a universal culture plate that covers both Gram Positive & Gram Negative bacteria. This plate is a Multi-Purpose Culture Plate.

Antibiotic Options

Amikacin

Highly effective against Gram Negative Bacteria, such as *Klebsiella* & *Pseudomonas.* Although, an excellent weapon against certain Gram Positive bacteria.

Gentamicin

Highly effective against Gram Negative Bacteria, especially *Pseudomonas. A*lthough some cross-over effects against Gram Positive Bacteria, a universal antibiotic although, it can be somewhat harsh if not buffered properly with $NaHCO_3$ & saline.

Penicillin

Effective against Gram Positive Bacteria excellent on all forms of *streptococci* an inexpensive & commonly used antibiotic.

Polymixin B

Effective against Gram Negative Bacteria especially the dreaded bug *Pseudomonas*, somewhat of a has been antibiotic.

Streptomycin

Effective against a wide variety of aerobic Gram Negative and some Gram Positive Bacteria

Ceftiofur (Naxcel® equivalent)

Highly effective against Gram Positive & Gram Negative Bacteria An all around super antibiotic.

Timentin (Ticarcillin & Clavulanic Acid)

Effective against Gram Positive & Gram Negative Bacteria excellent for *Pseudomonas* or *Staphylococci* bacteria varieties. Timentin® is extremely effective yet sensitive for use in semen extenders.

These guidelines are generalized and treatment should be based from the sensitivities of a uterine culture.

Laboratory Tips

***All laboratory equipment requires a specific voltage current. Voltage decline can easily occur if an improper wire gauge is used for the main underground/above ground electrical feeds. This is a very common shortcut that electricians are guilty of which saves them money and costs you the early replacement of your valuable laboratory equipment.

***Make sure that all of the receptacles in your lab, generates the exact voltage, a simple voltage meter will divulge the exact voltage that each receptacle is generating. Its better to have a greater output of voltage than reduction ie: 110 volts minimum is required to run your laboratory equipment as specified in the user guides.

***Its always a good idea to clean and prepare your lab equipment prior to the start of the breeding season. This only extends the life and requires little time.

***A can of Endust® Air Duster works very well, easily blowing out any of the accumulated dust from the previous breeding seasons. This simple annual tune up will add years to the life of all your laboratory equipment.

***A indoor/outdoor humidistat works extremely well to register the internal temperature and humidity in your incubator. It also keeps an extremely accurate record of the laboratory temperature. We offer an inexpensive humidistat in our catalog item **#191**.

***The *Quick-Temp* digital probe thermometer is by far the best inexpensive thermometer on the market. Works great for setting an AV, semen extender temperature and/or thawing frozen straws catalog item **#175**.

***It is suggested to thoroughly clean your A.V. with a non-abrasive brush such as our Scrubber Brush **#158** as well as a gentle antibacterial cleaner such as the A.V. Scrubber Gel **#159**, rinse thoroughly and hang in a storage cabinet.

The old theory of storing you're A.V.'s in 99% alcohol is not conducive to a long productive life of the gum rubber. For best bio-security, we suggest that you have cleaned you're A.V. and it has begun to air dry, place an A.V. Protector Cap over the end and hang on one of our ***Kept Safely™ A.V. Hangers **#172** in a clean dark place such as a specific A.V. Cabinet.

***Proper storage of your A.V. after the breeding season is very important. For best long term results, we suggest that you empty the water completely allowing the water to drain, leaving the valve open hanging down for 2 - 3 days. Replace the valve and put about 1 - 2 lbs of air through the air valve, cover the A.V. with baby powder, inside and out and place in a plastic bag and store in the freezer until next breeding season.

***Placing mares under artificial light 60 to 90 days before the start of your projected first breeding, will induce your mare to cycle much earlier. The artificial light should be equivalent to thirty (30) candle power strength at eye level throughout your entire holding facility.

***The use of **Regumate**® in your breeding program can be a very useful tool in order to group cycle your broodmares for either a scheduled foaling or embryo transfer.

***Prior to your final scrub and before insemination, make sure that you swab out the clitoral fascia, expelling out the tan colored debris. This area naturally collects urine salts as well as other impurities expelled from the urinary/reproductive tract. If the area is not clean, as you enter the vagina, you drag your "sterile" sleeve over this area which opens the door to an uterine infection.

Most people never take time to consider the point that once you open a tube of sterile lubricant, even if you try to be as aseptic as possible, the tube become a fantastic medium for growing bacteria. Our ***Next Generation*®* ***One Shots™ were designed to eliminate this situation!

***Before you insert the pipette, its always a good habit to palpate the cervix for any tears and/or thinning that may be present. If you inseminate with your right hand, simply place your thumb inside the cervix and your index finger outside and thread the cervix between your fingers clockwise. Mark any changes that appear to be abnormal as if you were looking at a clock. Torn or incompetent cervix will usually fail once the foal begins to grow and enter the third trimester of pregnancy.

***Oxytocin (P.O.P.) is a great tool for use in pre-breeding as well as post breeding therapies.

As a Pre-Breeding Tool:

Give two (2) hours prior to insemination. This will expel any excess fluid that may be present in the uterus that could contaminate and disrupt the quality of the semen post insemination.

As a Post-Breeding Tool:

Give two (2) hours post insemination. This will assist in the expulsion of any excessive seminal debris that may still be present in the uterus. Your veterinarian can inform you as to the correct dosage for the age & weight of your broodmare. I believe this is one of the most effective tools that can be easily implemented in your daily breeding program.

Foaling Quick Tips

Heart Rates - Normal: 60 to 110 beats/minute

Respiration Rates:

1 Hour Post Foaling: 70 - 90 breathes/minute

1 Week of Age: 25 - 60 breathes/minute

Rectal Temperature: 100F - 102.5F

***To calculate the foaling date of your mare on the normal 340 day gestation, calculate the following:

1. Take the mares last breeding date
2. Add five days
3. Subtract 30 days

***Make sure your booster shots are given 45 days prior to the date due to foal. This will maximize the titer giving your foal a huge jumpstart!

***Bring your foaling mares into the barn at night, 25 days before their due date. If a mare foals prior to 25 days, it is unlikely the foal will survive without a huge medical expense.

***Make sure that your foaling barn is quiet at night and your mares are comfortable with your night watch person. If your mares begin to foal during the day or in an irregular pattern, look first to your night watch person – Shhhhhhh!

***Castlicks (suturing the vulva closed to prevent wind sucking in the vagina) is an extremely useful tool with sub-fertile broodmares. Most trainers are having them completed before a filly even goes in to training. It is very important to make sure your broodmare does not have a castlicks intact, they will tear open and present you with a real mess post foaling as well as a broodmare who may need re-constructive surgery just to get re-bred!

***A broodmare will relax the following parts of her body just before foaling.

1. Muscles and ligaments along her rump.

2. Her vulva will begin to lengthen.

3. Her teats will fill while becoming very blackened and shiney the closer she gets to foaling.

4. Record her daily changes as she progresses towards her foaling due date. This information will enable you to predict future foaling patterns.

5. If you have a night watch person on staff and most of your mares foal during the day, your night watch person is either too noisy or they are making your mares too restless to comfortably foal.

***Always keep good breeding & foaling records. This will pay off in spades in the future.

***Always perform tests for the passive transfer of IgG

1. On farm IgG site tests work very well #420

2. Use a certified Equine Colostrumeter. #423

3. Send blood out to a commercial lab.

*****Thank GOD each and every time you get a live foal*****

*** Management of your recipient herd is absolutely vital to the success of any embryo transfer program. Tasks that need to be closely observed are:

1. Lighting program to begin at least 75 days prior to your first desired breeding date and 15 days before your donor herd.

2. Pre-season reproductive examinations including:

a. Uterine culture and/or biopsy

b. Rectal & vaginal palpation exam

c. Ultrasound examination of the entire reproductive tract

***The management of the donor herd should be on the same track as the recipient herd, as listed above.

***When flushing, it is best to use a large volume flow and return tubing set up. This accomplishes two (2) things:

1. Completely floods and hyper extends the uterus quickly allowing for less endometrium irritation.

2. Your opportunity to flush an embryo that that may be trapped in the tip of the uterine horn or has not dropped completely into the uterus from the oviductal regions will be greatly increased.

***When scheduling your embryo flushes, day seven (7) embryos (blastocysts) transport much better than do day six (6) or eight (8).

***If you flush a day seven (7) embryo, it is suggested that you transfer it into a day five (5) or early day six (6) post ovulation receipt mare.

***Always attempt to keep a two day interval between the donor mare and the recipient mare. This allows an extra two days for the embryo to acclimate into the new uterine environment.

***Embryo contact and continual movement within the uterus, sends signals to the pituitary gland that the mare is pregnant. If you transfer a day seven (7) embryo into a day seven (7) post ovulation mare, you'll automatically reduce the natural intra-uterine contact and dramatically increase the chance of complete embryonic rejection. Strongly consider placing 7 or 8 day embryos in 5 day post ovulation mares.

*** Always keep your recipient and donor herds on the same vaccination schedule. This reduces the opportunity of embryonic rejection due to potential vaccine reactions.

*** Once you establish a group of mares whether it be your donor herd or receipt herd, do not introduce new members for any reason

***Breeding with frozen semen requires a very narrow window for insemination, six hours prior to or post ovulation. The following protocol should be followed closely in order to achieve success, this procedure is expensive as well as extremely time consuming.

1. Reproductive examinations every 8—12 hours to follow the follicle right up to ovulation.
2. An ultrasound must be used in order to pinpoint the progress, growth as well as exact ovulation.
3. Insemination must occur within the "Golden Period" of a **maximum** of 6 hours post ovulation.

***It is important to know the exact inventory in your nitrogen tank. In each goblet the individual canes should be marked for quick and easy access. Using a (6) six column accounting pad works very good in order to maintain a good purchase/usage inventory.

***When handling frozen semen, it is extremely important that the goblet not be kept out of the nitrogen for longer than 20 seconds at a time. This alone will decrease the risk of damaging the other straws of semen that reside in the goblets held in the aluminum cane.

***It is best to have an experienced technician place the semen at the tip of the uterine horn. This is achieved using a 30" flexible pipette and directing the pipette into the desired uterine horn via rectal palpation. This two-step process that begins vaginally and then completed via rectal manipulation, is a very difficult procedure to properly accomplish.

***Post ovulation infusions are an extremely important tool when using frozen semen. In each insemination dose, the sperm cells are spun down into a tiny pellet by a centrifuge. The concentration of those sperm cells are so great, that the uterus will profoundly reject the dead cells that were lost during the freeze/thaw process. A warm uterine lavage, with saline will normalize the uterine pH as well as expel excess debris.

***Using frozen semen has many benefits!

1. Virtually eliminates the expense of semen transport
2. Your breeding options are open 7 days a week
3. Allows for the control of planned foalings

***Using frozen semen requires a tightly managed breeding program utilizing an ultrasound. Frozen semen will not be for every operation. Although, it will open up many options that in the long run can and will improve the genetics of a breed.