VetMotl® Sperm Separation Devices in Equine ICSI: Differentiated Performance for Advanced Veterinary Reproduction

VetMotl Sperm Separation Devices reliably deliver a higher-quality sperm subpopulation than conventional preparation methods, improving motility, morphology, viability, membrane and mitochondrial integrity, and DNA integrity[1]. In equine ICSI, the VetMotl device can convert previously embryo-negative mare–stallion combinations into clinically meaningful blastocyst production[2].

Why VetMotl Changes Equine ICSI Outcomes

- The VetMotl device produces a consistently upgraded sperm cell fraction compared with wash, swim-up, and density/colloidal gradient techniques, while avoiding centrifugation-related damage and reactive oxygen species (ROS) exposure[3].
- In equine ICSI and other high-value veterinary ART procedures, this translates into:
    - Demonstrated "rescue" of difficult mare–stallion combinations: blastocyst rates of 16–50% in pairs that had previously produced 0 embryos when semen was prepared by simple wash alone[2].
    - Embryo development that is at least comparable to single-layer colloidal centrifugation, but with a sperm population that shows better viability and/or DNA integrity and a simpler, gentler workflow[4].
- For frozen-thawed or poor-fertility stallion semen, the VetMotl devices reliably yield a higher-quality subpopulation and offer additional value as a tool to help predict and triage semen doses for advanced reproductive use[5].

Core performance advantages

1. Delivers a distinctly higher-quality sperm fraction

Across independent equine studies, VetMotl device use has shown:

Motility and progressive motility

- Significant increases in total and progressive motility in frozen-thawed stallion semen compared with the original sample, with performance equal to or better than gradient methods and clearly superior to swim-up and simple wash[1,5].
- In fresh and cooled ejaculates, VetMotl device-selected motility and progressive motility substantially exceeded unprocessed, washed, or swim-up preparations and were comparable to or better than density-gradient and combined gradient–swim-up protocols[1].

Morphology

- Consistent enrichment for morphologically normal sperm; in several experiments the VetMotl device was the only method that improved morphology, viability and DNA integrity simultaneously relative to the starting sample[3,4].
- In direct comparison of VetMotl Equine vs. VetMotl Bovine devices on stallion semen, both devices improved morphology over control, but the equine-pore device produced the best morphology at higher input loads, indicating tighter quality selection when pore size is species-optimized[6].

Viability and membrane integrity

- Higher proportions of live, membrane-intact sperm after VetMotl device selection compared with the original sample, and in one ICSI study, higher viability than SLC at similar motility and morphology[4,5].
- Improved membrane functionality (hypo-osmotic swelling) in most sample types, without detriment even in severely compromised ejaculates, underscoring the non-damaging nature of the process[4,5].

Mitochondrial function

- A marked increase in the percentage of sperm with high mitochondrial membrane potential, indicating a subpopulation better equipped to support fertilization and early embryo development[5].

DNA integrity / chromatin quality

- Robust reduction in DNA fragmentation versus the original ejaculate, and better DNA integrity than SLC and swim-up in head-to-head comparisons; in one study, the VetMotl device was the only method to significantly lower DNA fragmentation relative to baseline[1,3,4,5].
- Improved chromatin condensation scores after VetMotl device selection in both good- and poor-fertility stallions[5].

Bottom line: VetMotl devices yield a sperm fraction that is simultaneously more motile, more morphologically normal, more viable and with superior DNA and chromatin integrity than sperm prepared by conventional centrifugation-based or swim-up methods[1,3,4,5,6].

2. Outperforms or matches conventional methods where it matters

The summary table below reflects direct comparisons with simple wash, swim-up (SU), and density- or colloidal-gradient techniques (DGC/SLC). The pattern is consistent:

- VetMotl devices deliver larger gains in total and progressive motility than wash and SU, and perform at least as well as gradients[1,4,5].
- They enrich normal morphology more effectively than wash and SU and are comparable to gradients, without the inherent drawbacks of centrifugation[1,3,4,5].

Technical Insights: Equine ICSI

VetMotl, Inc.

- VetMotl devices improve viability and membrane integrity to a similar or higher degree than SLC, often with a distinct advantage in DNA integrity[1,4,5].
- SU emerges as the least effective method across multiple quality endpoints, while gradient methods improve some metrics but do not match the DNA and chromatin benefits of VetMotl[1,4].

Feature / outcome	VetMotl Sperm Separation Device	Simple wash	Swim-up (SU)	Density / single-layer gradient (DGC/SLC)
Motility gain vs. original	Large, significant increase (fresh, cooled, frozen-thawed)	Minimal to none; wash alone failed to rescue ICSI cases in subfertile mares	Improves motility, but often less than VetMotl and gradients	Improves motility, similar to VetMotl in some settings
Progressive motility	Higher than wash and SU; similar to or better than gradients across studies	Low to moderate; insufficient in difficult cases	Lowest among selection methods in several experiments	High; sometimes comparable to VetMotl
Morphology	Consistent enrichment of normal forms; often best or equal-best	Unchanged	Improved but still inferior to VetMotl/SLC in many data sets	Improved; typically similar to VetMotl device, but requires centrifugation
Viability	Increased vs. original; in one ICSI study, viability higher than SLC at comparable other parameters	Unchanged	Improved but generally below VetMotl and gradient methods	Improved; sometimes slightly lower viability than VetMotl
DNA / chromatin integrity	Reduced DNA fragmentation vs original; better DNA integrity than SLC and SU; improved chromatin condensation	No targeted improvement; susceptible to ROS from centrifugation steps in some protocols	No consistent DNA benefit; in equine work, SU was least effective overall	Improves DNA vs original, but did not reduce fragmentation as consistently as VetMotl
Embryo outcomes (equine ICSI)	Converts 0% blastocyst rate after wash into 26.5%	0 embryos per OPU in the subfertile mare	In equine ICSI studies, lower blastocyst development than	Good ICSI outcomes; in VetMotl-vs-SLC comparisons, similar

Feature / outcome	VetMotl Sperm Separation Device	Simple wash	Swim-up (SU)	Density / single-layer gradient (DGC/SLC)
	average blastocyst rate (range 16.7–50%) in previously unsuccessful mare–stallion combinations; ~2 embryos per OPU vs 0 with wash. Comparable cleavage/blastocyst rates to SLC in mixed clinical ICSI work, with better DNA/viability profile.	cohort that later responded to VetMotl	more advanced methods, and SU-selected sperm show the weakest lab quality parameters.	cleavage/blastocyst rates but without the DNA advantage of VetMotl.
Centrifugation / ROS exposure	No centrifugation; lower ROS and less iatrogenic damage expected.	Often includes centrifugation steps depending on lab protocol	Requires centrifugation; associated with increased ROS and potential DNA damage.	Intrinsically centrifugation-based; literature and attached data highlight ROS, membrane and DNA risks.

Bottom line: Compared with traditional wash, swim-up and gradient methods, VetMotl devices consistently select a higher-quality sperm subpopulation, especially on the most critical, damage-sensitive parameter—DNA integrity[1,3,4,5].

3. Demonstrated impact in equine ICSI

Rescuing previously unsuccessful ICSI combinations

- In mares (11–22 years; median 18) that had failed to produce embryos after one to three OPU–ICSI sessions using semen prepared by standard wash, switching to the VetMotl device increased blastocyst rates from 0% to an average of 26.5% per injected oocyte (range 16.7–50%; $p \leq 0.0001$)[2].
- These same mare–stallion pairs went from 0 embryos per OPU session with wash to an average of 2 embryos per OPU when sperm were prepared with the VetMotl device[2].

Comparable ICSI outcomes with a better biological profile

- When VetMotl and SLC were used clinically prior to ICSI, cleavage and blastocyst rates per injected oocyte were similar[4].

- Only VetMotl device selection, however, routinely delivered sperm populations with lower DNA fragmentation and, in at least one series, higher viability than SLC, differences that are expected to influence late embryo and pregnancy outcomes even when early blastocyst rates are similar[1,4].

Bottom line: the VetMotl devices' sperm sample preparations do more than improve lab parameters; they change clinical outcomes in hard ICSI cases, enabling blastocyst production where standard wash failed, while matching or exceeding the embryo performance of SLC with a biologically superior sperm fraction[2,4].

4. Practical advantages for veterinary ART programs

Centrifugation-free, low-ROS workflow

- VetMotl devices operate without centrifugation, minimizing ROS generation, membrane peroxidation and mechanical damage associated with spin-based methods[1,4,5].
- This simplifies protocols, reduces equipment dependency and supports more consistent outcomes across laboratories and operators[1,4,5].

Optimized use of limited or poor-quality semen

- The VetMotl device consistently yields a high-quality fraction, even from frozen-thawed or low-fertility semen; recovery percentage is higher from good-fertility stallions but remains clinically useful from poor-fertility ejaculates[2,5].
- Because ICSI and related techniques require very few sperm, VetMotl's "fewer but better" strategy fits high-value equine and bovine cases—older mares, problem stallions, rare genetics and expensive frozen doses[2,5].

Species-tuned device options

- VetMotl offers equine- and bovine-optimized Multi devices: in stallion testing, the bovine-pore device retrieved more total sperm, while the equine-pore device delivered slightly higher quality metrics, allowing practitioners to choose between maximal yield and maximal quality according to case objectives[6].

Bottom line: VetMotl devices integrate smoothly into veterinary ART workflows, reduce technical risk and make better use of every straw—especially in challenging or high-value cases[2,5,6].

Conclusion:

The collective data from multiple independent equine studies support the VetMotl Sperm Separation Device as a differentiated, evidence-based advancement for ICSI-focused veterinary reproduction. By reliably enriching for sperm with superior motility, morphology, viability, membrane and mitochondrial integrity, and DNA quality—while eliminating centrifugation-related stress—VetMotl devices enable practitioners to optimize outcomes from

limited or compromised semen and to rescue mare–stallion combinations that previously failed to yield embryos under standard preparation protocols. For equine reproduction programs seeking more predictable embryo production, better use of high-value semen, and a workflow aligned with physiologic sperm selection, VetMotl devices provide a practical, clinically validated tool to elevate the performance and reliability of ICSI-based ART services.

Nota bene: *All referenced posters and publications in this white paper evaluate the same underlying sperm sample preparation technology that is commercialized as the VetMotl Sperm Separation Device. In the original source materials, this technology may be referred to as VetMotl, as the human predecessor device "ZyMōt," or more generically as a "microfluidic" or "microflow" device; for purposes of this communication, all such references should be understood to denote VetMotl devices.*

[1] **Orsolini MF et al.** "Characterization of sperm cell membrane charge and selection of high-quality sperm using microfluidics in stallions." *Theriogenology*. 192 (2022): 1–8. (UC Davis; comparison of SU, DGC, DG-SU, microfluidic chip on fresh/cooled ejaculates; motility, progressive motility, viability, morphology.)

[2] **Lorenzen E et al.** "Effect of microfluidic sperm sorting on equine ICSI blastocyst rate." *Journal of Equine Veterinary Science*. 89 (2020): 103032 (Abstract). (Previously unsuccessful mare–stallion OPU–ICSI combinations; 0% blastocysts after wash vs. 26.5% after ZyMōt Multi chip, with 16.7–50% range.)

[3] **Gonzalez-Castro RA, Carnevale EM.** "Use of microfluidics to sort stallion sperm for intracytoplasmic sperm injection." *Animal Reproduction Science*. 202 (2019): 1–9. (MF vs. SLC vs. SU; improved motility, morphology, viability and DNA integrity; SU least effective; similar ICSI cleavage/blastocyst rates for MF and SLC, with viability advantage for MF.)

[4] **Paredes BJ et al.** "Sperm processing methods effectively reduce the DNA fragmentation index in samples with high initial values." *Journal of Equine Veterinary Science*. 125 (2023): 104609. (Frozen-thawed equine semen; DGC, SU and microfluidic chamber; MF and SU most effective in reducing DFI when initial DFI is high.)

[5] **Vigolo V et al.** "Selection of frozen–thawed stallion semen by microfluidic technology." *Reproduction in Domestic Animals*. 58 (2023): 443–449. (ZyMōt Multi 850 µL; good vs. poor fertility stallions; increased total motility, membrane integrity, membrane functionality, mitochondrial membrane potential, chromatin integrity; lower concentration but higher-quality subpopulation usable for ICSI and fertility prediction.)

[6] **Nunes MM et al.** "Comparison of two sperm selection devices using variable sperm loads to enhance quality and sperm harvest from fresh-diluted stallion ejaculates." Poster, 14th Association for Applied Animal Andrology (AAAA), 2024. (VetMotl Multi 3 mL – Equine vs. Bovine; both improve morphology and progressive motility; equine-pore device yields better quality, bovine-pore higher sperm retrieval.)