



EquiPro® - Preservation extender medium for fresh and cooled stallion semen

EquiPro® is a defined and standardized extender for the preservation of fresh and cooled stallion semen. It is comprised of several tested sugars and buffers and is available without antibiotics or with antibiotics to control bacterial growth. EquiPro's® unique formula includes caseinates, derived from various fractions of milk casein, and highly purified by high-end processing techniques. Caseinates have proven to be the most effective for the preservation of stallion semen due to the protection they provide to the sperm membrane.

EquiPro® is available in powder form or as a ready to use liquid. Liquid EquiPro® is a sterile, milky-white solution that only requires a simple warming step prior to use. Powdered EquiPro® is a white powder, which must be dissolved in purified water prior to use.

EquiPro® is suitable to use for centrifuged and non-centrifuged semen.

EquiPro® ApX^{2™} is the newest addition to the EquiPro® product line and contains a powerful fertility enhancing antioxidant called Ergothioneine. Ergothioneine is naturally present in stallion seminal plasma and works to protect sperm cells from oxidative stress. When blended with EquiPro®, ApX^{2™} Ergothioneine works to delay programmed cell death (apoptosis).

EquiPro® ApX^{2™} is available with different antibiotics to meet the individual needs of each stallion.

EquiPro® in scientific studies

Traditional milk and egg yolk based extenders consist of a range of biological substances that can suffer from batch-to-batch variation, thus increasing the risk of microbiological contamination. In contrast, EquiPro® extenders contain defined and highly purified proteins (DMP) manufactured in standardized batches. The end result is a product that is superior to skim milk extenders when semen is stored at 5°C (Aurich 2006). In a comparative study, Pagl et al. (2006) concluded that semen preserved with EquiPro® (DMP) showed better results for motility and membrane integrity over a storage period of 72 hours when compared to skim milk extender.

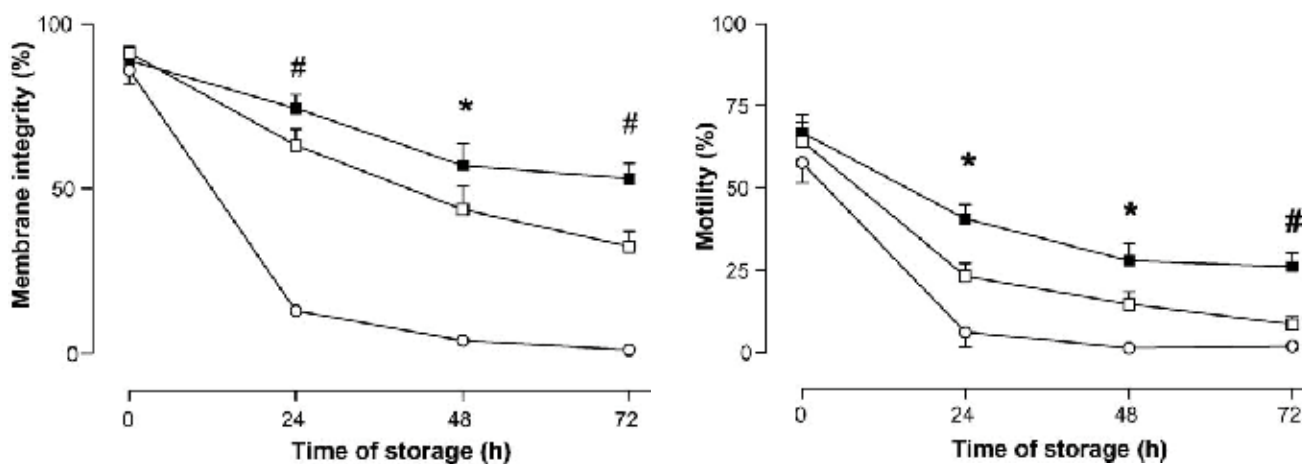


Diagram 1/2: Percent of membrane-intact spermatozoa and motility in non-centrifuged semen diluted in either PBS (o) or defined milk protein (DMP) extender (□) and in centrifuged semen diluted in DMP extender (■) during storage at 5°C for 72 h. Source: Pagl et al. (2006)

Significant differences between extenders became very evident after semen was stored for more than 24 hours. The authors concluded that there is a potential advantage to EquiPro® when semen is stored over a period of 2 days. Semen centrifuged and extended with EquiPro® showed

almost identical values for motility and membrane integrity after 48 hours and 72 hours, suggesting a benefit of centrifugation when semen has to be stored for up to 3 days.

Another recent study conducted by LeFrappier et al (2010) also showed that EquiPro® has excellent performance when extended up to 3 days. Through out the study EquiPro® was in the top group showing the best results for both total motility and progressive motility over a storage period of 72 hours ($P < 0.05$). Semen extended with EquiPro® was also among the top of the test groups for the CASA parameters including curvilinear velocity, straight-line velocity, and curvilinear distance ($P < 0.05$) (LeFrappier 2010).

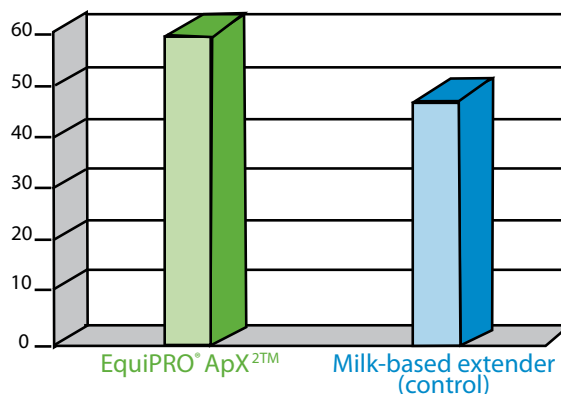
EquiPro® ApX²™

Motility and fertility of semen stored at +5°C for 24 hours in EquiPro® ApX²™ was tested in preliminary studies with 352 mares (ponies and horses). These studies showed an advantage in fertility and no disadvantage in motility when semen was extended in EquiPro® ApX²™ compared to basic milk-based extenders. Based on these preliminary data, it was hypothesized that semen extended in EquiPro® ApX²™ for storage under controlled cooled conditions would have higher fertility when compared to semen extended in a basic milk-based control extender.

Some facts about EquiPro®

EquiPro® is available in different formulations and package sizes:

- as powder with and without antibiotics
 - 100 ml package
 - 1 liter package
- in liquid formulation, 100 ml ready to use



Pregnancy rates in mares inseminated with shipped cooled semen extended in EquiPro® ApX²™ and the control milk-based extender. The difference was statistically significant at a level of $p=0.05$

Minitube provides detailed information about the ingredients of EquiPro® and the type and amount of antibiotics used. There is scientific evidence that not only bacteria but also some antibiotics may have harmful effects on spermatozoa (Aurich & Spengler 2007), with individual variability among stallions. It is, therefore, a good option in certain cases to choose an extender without antibiotics or with a different selection of antibiotics: e.g. for stallions that are categorized as “bad coolers”. Centrifugation can also improve the properties of cooled semen.

Strict quality control of the production and sterilization process eliminates the risk of mold growth contamination in liquid EquiPro®. Due to Minitube’s manufacturing process, no extra fungicides are required to be added unlike other commercially available liquid stallion semen extenders. Fungicides can be harmful to the overall fertility of the sperm. According to Pagl et al. (2006), antibiotics, fungicides and generally any extender additive should be avoided unless it protects the sperm. Spermatology research shows that in other species, there is evidence that antifungal agents in semen extenders tend to be highly spermicidal, for example to bull and rabbit sperm (Foote, 2002).

EquiPro® liquid extender is packaged and delivered in a glass bottle with a secure cap. Glass is inert making it the ideal container for packaging the extender. Plastic containers on the other hand, may release plastic molecules into the extender fluid, contaminating the extender and potentially harming sperm. Once the EquiPro® liquid is opened or the EquiPro® powder is mixed with water, the extenders can be kept for up to 3 days in the refrigerator at +5°C.

In practice and in science, the temperature range of +4-6°C is considered to be the optimal storage temperature to preserve both motility and fertility of equine semen over 24 hours and more (Varner et al., 1989). Shipped cooled semen has grown in popularity because semen shows almost no loss of fertility when stored at +5°C for about 24 hours (Aurich 2005). EquiPro® trials also support this statement that the best results for 24 hour old semen are generally achieved with storage temperature at +5°C (Price et al. 2008, Pagl et al. 2006).

Conclusion

EquiPro® is an extender for stallion semen and ideal for the conservation of sperm at +4°C to +6°C for up to 3 days. In numerous studies as well as in the field, EquiPro® has proven to provide consistent results with a wide range of stallions.

Based on the proven EquiPro® formulation, Minitube has also recently introduced EquiPro® CryoGuard™, a series of semen extenders for the long term cryopreservation of stallion semen.

References

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