

## Recipient of the 2024 IETS Pioneer Award: Dr Edward Squires

Dr Squires grew up on a small farm in Morgantown, West Virginia, the home of West Virginia University. At age 11, he got his first job working at a stable, so it was only natural that he stayed at home and continued to work with horses and attend the University. After obtaining a BS in Animal Sciences he was solicited to work on a Master of Science degree under the guidance of a prominent reproductive biologist, Keith Inskip. It was Dr Inskip that encouraged him to attend the University of Wisconsin for a PhD, under the mentorship of a previous pioneer award winner, O. J. Ginther. Dr Ginther had several PhD students working at that time on various aspects of mare reproduction. Dr Squires was assigned to study the follicular and luteal development in pregnant mares. One of his early papers demonstrated that the primary CL in the pregnant mares was viable to 120–150 days of gestation and that eCG rescued the primary CL from regression. Squires went on to demonstrate the role of the ovary and placenta in pregnancy maintenance. In 1976, Squires took a position at Colorado State University and joined the team at the Animal Reproduction Laboratory. He remained as part of that productive group for 33 years. In 1981 he published a novel paper on surgical and non-surgical equine embryo transfer. His team went on to develop the technics for maintaining the viability of embryo stored at 5°C. Through the training of veterinarians from all over the country, the practice of collecting and shipping embryos to recipient stations became commonplace. One other area of great interest was the application of the oral progestin altrenogest, for managing the mare's cycle. Squires and his students published numerous papers on the use of Altrenogest in vernal transition as well as cycling mares. Other studies showed the value of Altrenogest for pregnancy maintenance in broodmares and recipients.

With the help of Dr Terry Nett, Squires investigated the seasonal changes in hypothalamic GnRH, GnRH receptors and pituitary LH and FSH. Squires also investigated the use of GnRH for ovulation control and follicular development.

Also, during those early years at CSU, Squires took the opportunity to work with Bill Pickett, the famous stallion reproductive physiologist. They published on many aspects of stallion management, such as factors affecting sperm output and sexual behaviour. His work also included identifying the best extenders for cooling and freezing semen. With the help of Dr Amann, they determined the proper cooling curve



needed for maintaining viability of semen cooled to 5°C. Jim Graham, at CSU was also instrumental in designing experiments to evaluating the use of liposomes and cholesterol in semen extenders.

Squires is best known for his work in developing assisted reproductive technics for the mare and stallion. This technology was transferred to the veterinarians and breeders through short courses. These 3–5-day courses were started in the early 1970 and continue to the present. Thousands of breeders and veterinarians were taught the technics of AI with cooled and frozen semen, embryo transfer, ultrasound and cryopreservation of oocytes, sperm, and embryos.

Obtaining funding for equine research is a continual problem. A turning point in the equine research program at CSU occurred in the late 1990s. This was the creation of a privately funded research program, called the Preservation of Equine Genetics (PEG). The emphasis was equine Assisted Reproductive Technology (ART), not genetics, and several million dollars were given to CSU by breeders. These funds were used competitively to fund graduate students and faculty doing ART. It was through these funds that scientists such as Drs Carnevale, McKinnon, Seidel, Nett, Graham were able to

participate in the PEG program. Out of that program came the development of technologies such as oocyte collection and transfer, superovulation, cooled and vitrified embryos and sexed semen. It was very fortunate that Drs Seidel, Graham and Nett were all at CSU during the majority of Squire's career. At that time, Dr George Seidel had developed one of the largest research and commercial programs in bovine ET in the world and Squires was fortunate to have him involved in all the equine embryo projects. XY, the semen sexing company also had its start at CSU and they were able to publish the birth of the first horse foals born from mares bred with sexed semen.

Superovulation of mares was also a passion of Squires. He initially tested the use of crude pituitary equine FSH for superovulation and then went on to demonstrate that a more purified pituitary FSH could be used to recover two to four embryos from FSH-treated mares versus 0.5 from non-treated mares. He also evaluated the use of recombinant FSH for superovulation of mares and is currently working with Pablo Ross and Jan Roser to produce a recombinant equine FSH for the equine market.

In collaboration with Paul Loomis at Select Breeders, Squires published several papers to identify the barriers for the use of frozen semen. These included the proper sperm numbers and frequency of AI, and mare management needed for maximum fertility with frozen semen.

In 2008, Squires moved to the Gluck Equine Research centre at the University of Kentucky. With the assistance of Mats Troedsson and Barry Ball they were able to get support from the Thoroughbred industry to study pregnancy losses in mares and post-breeding endometritis. The measurement of cytokines in resistant and susceptible mares provided insight into the possible treatment for endometritis.

He has published 339 articles in refereed journals and 20 chapters in textbooks and one of the Editors for the very popular text, 'Equine Reproduction' second edition by McKinnon, Squires, Varner, and Valla. He is co-author of a book titled 'Equine Embryo Transfer' by PM McCue and El Squires (2015).

Squires has attended nearly all of the IETS meetings since 1982 and has been an invited speaker at four of the annual meetings. He served on the board of governors from 2005 to 2007. Squires served on the local organising committee twice, 2008 and 2016. Other areas of service include serving on the data retrieval committee for nearly a decade and the IETS Foundation board. He also served as a reviewer for abstracts on many occasions and was the chair of the stallion section for the horse IETS pre-conference in Paris in 2015. His vision is to continue to have a balance in the society where basic scientist, translational scientist, practitioners, and industry leaders can participate in the exchange of knowledge. One of his goals is to see that new technology is translated so that it can be used effectively in agriculture and biomedical science. Squires has extensive experience in teaching, research, clinical reproduction, the horse and veterinary industry and fund raising from the horse industry for research.

He has lectured extensively nationally and internationally to veterinary and scientific groups and horse breeders in nearly 40 foreign countries. He feels strongly that IETS should be a source of information for the practitioner and governing bodies and should be a leader in presenting new technologies.

Squires has received numerous awards including to be inducted into the Equine Research Hall of Fame, Distinguished Alumni Award College of Agriculture West Virginia University, George Stubb Award from the American Association of Equine Practitioners, Honorary member of the College of Theriogenology, honorary vice president American Quarter Horse Association, past president of the Equine Science Society and chair of the International Symposium on Equine Reproduction and most recently Honorary Chair of the XIII International Symposium on Equine Reproduction.

He is a father of four sons and has seven grandchildren all living in the west within two hours of Denver. He and his wife Norma enjoy the farm they have in Colorado and spend time with the kids riding horses and camping. He currently has a consulting business and serves as a large animal technical specialist for Vetoquinol and research advisor for Select Breeders service.

## References

- Canisso IF, Ball BA, Esteller-Vico A, Williams NM, Squires EL, Troedsson MH (2017) Changes in maternal androgens and oestrogens in mares with experimentally-induced ascending placentitis. *Equine Veterinary Journal* **49**(2), 244–249. doi:10.1111/evj.12556
- Carnevale EM, Squires EL, McKinnon AO (1987) Comparison of Ham's F10 with CO<sub>2</sub> or Hepes buffer for storage of equine embryos at 5C for 24 H. *Journal of Animal Science* **65**, 17. doi:10.2527/jas1987.6561775x
- Carnevale EM, Squires EL, Maclellan LJ, Alvarenga MA, Scott TJ (2001) Use of oocyte transfer in a commercial breeding program for mares with various reproductive pathologies. *Journal of the American Veterinary Medical Association* **218**, 87–91. doi:10.2460/javma.2001.218.87
- Eldridge-Panuska WD, Caracciolo di Brienza V, Seidel GE Jr., Squires EL, Carnevale EM (2005) Establishment of pregnancies after serial dilution or direct transfer by vitrified equine embryos. *Theriogenology* **63**, 1308–1319. doi:10.1016/j.theriogenology.2004.06.015
- Fedorka CE, Scoggin KE, Boakari YL, Hoppe NE, Squires EL, Ball BA, Troedsson MHT (2018) The anti-inflammatory effect of exogenous lactoferrin on breeding-induced endometritis when administered post-breeding in susceptible mares. *Theriogenology* **114**, 63–69. doi:10.1016/j.theriogenology.2018.03.017
- Hart PJ, Squires EL, Imel KJ, Nett TM (1984) Seasonal variation in hypothalamic content of gonadotropin-releasing hormone (GnRH), pituitary receptors for GnRH, and pituitary content of luteinizing hormone and follicle-stimulating hormone in the mare. *Biology of Reproduction* **30**, 1055–1062. doi:10.1095/biolreprod30.5.1055
- Imel KJ, Squires EL, Elsden RP, Shideler RK (1981) Collection and transfer of equine embryos. *Journal of the American Veterinary Medical Association* **179**, 987–991.
- Lindsey AC, Bruemmer JE, Squires EL (2001) Low dose insemination of mares using non-sorted and sex-sorted sperm. *Animal Reproduction Science* **68**, 279–289. doi:10.1016/s0378-4320(01)00165-8
- McCue PM, Patten M, Denniston D, Bruemmer JE, Squires EL (2010) Strategies for using eFSH superovulating mares. *Journal of Equine Veterinary Science* **28**(2), 91–96. doi:10.1016/j.jevs.2008.01.005

- Slade NP, Takeda T, Squires EL, Elsdon RP, Seidel GE Jr. (1985) A new procedure for the cryopreservation of equine embryos. *Theriogenology* **24**, 45–58. doi:[10.1016/0093-691x\(85\)90211-0](https://doi.org/10.1016/0093-691x(85)90211-0)
- Squires EL, McCue PM (2007) Superovulation in mares. *Animal Reproduction Science* **99**, 1–8. doi: [10.1016/j.anireprosci.2006.04.054](https://doi.org/10.1016/j.anireprosci.2006.04.054)
- Squires EL, Douglas RH, Steffenhagen WP, Ginther OJ (1974a) Ovarian changes during the estrous cycle and pregnancy in mares. *Journal of Animal Science* **38**, 330–338. doi:[10.2527/jas1974.382330x](https://doi.org/10.2527/jas1974.382330x)
- Squires EL, Wentworth BC, Ginther OJ (1974b) Progesterone concentration in blood of mares during the estrous cycle, pregnancy and after hysterectomy. *Journal of Animal Science* **39**, 759–767. doi:[10.2527/jas1974.394759x](https://doi.org/10.2527/jas1974.394759x)
- Weibel SK, Squires EL (1982) Control of the oestrous cycle in mares with altrenogest. *Journal of Reproduction and Fertility. Supplements* **32**, 193–198.
- Woodward EM, Christoffersen M, Campos J, Betancourt A, Horohov D, Scoggin KE, Squires EL, Troedsson MHT (2013) Endometrial inflammatory markers of the early immune response in mares susceptible or resistant to persistent breeding-induced endometritis. *Reproduction* **145**(3), 289–296. doi:[10.1530/rep-12-0452](https://doi.org/10.1530/rep-12-0452)