LIÈGE université
Médecine Vétérinaire

Description des profils de différents œstrogènes pendant la gestation de juments de différentes races par chromatographie liquide couplée au spectromètre de masse

Description of different estrogens profiles during the gestation of mares of different breeds by liquid chromatography coupled with the mass spectrometer

Ledeck Joy

Travail de Master de spécialisation présenté en vue de l'obtention du Master de spécialisation en sciences vétérinaires :

Internat clinique

ANNÉE ACADÉMIQUE 2020-2021

Contexte

Jument poulinière demi-sang de 18 ans

Gestante de 9 mois

A déclenché une lactation prématurée

Motif de consultation : suspicion de placentite

Contexte :

Examen clinique dans les normes

Examen spécial :

US : hétérogénéité de l’unité fœto-maternelle et augmentation de son épaisseur.

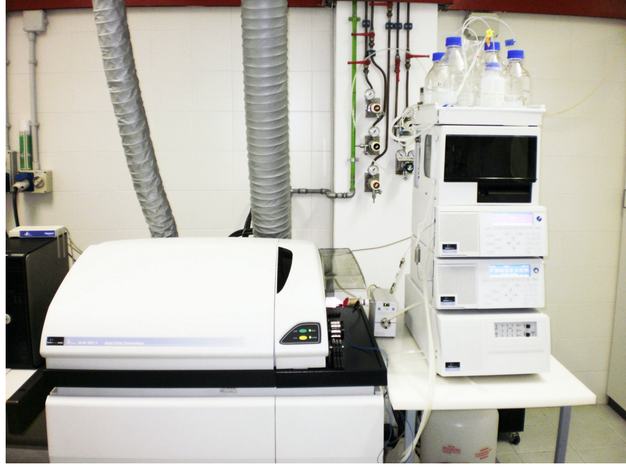
Peu invasif

Diagnostic plus aisé et précis à partir de 270 jours (Campos *et al.*, 2017)

Influence du stade de gestation, race mais pas l’âge

Placentite ≠ Epaississement: seules 60% des juments infectées expérimentalement montrent des signes échographiques de placentite.

Contexte :



Les propriétaires veulent une réponse immédiate

Mesure de l'E1S pour le diagnostic *(Satué et al., 2011 ; Esteller-Vico et al., 2017)*

Résultats : 300 ng/mL en LC/MS mais qu'en faire ?

Laboratoire 1 (RIA) :	Laboratoire 2 (ELISA) :
40 ng/mL	600-800 ng/mL

Cholesterol

↓

Prégnénolone

↓

17 -hydroxyprégnénolone

↳ Déhydroépiandrostérone

↳ Androsténédione → E1 → œstriol

↳ Androsténédiol → Testostérone → E2 → œstriol

↳ 7 Déhydroépiandrostérone → Equiline et Equilenine

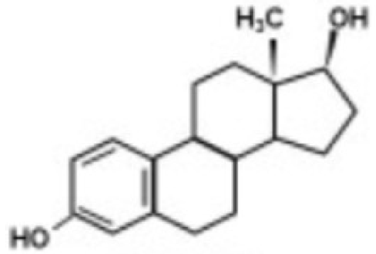
Buts de l’étude :

Evolution de la concentration en œstrogènes à partir de 4 mois de gestation jusqu’au poulinage à l’aide de la LC-MS/MS

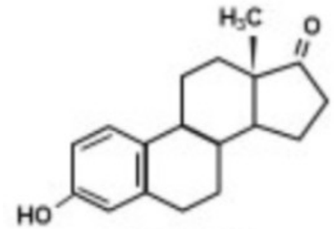
Courbe des concentrations en fonction du temps de l’E2, l’E1 et l’E1S chez des juments saines

Comparaison entre races

Comparaison entre juments saines et souffrant de placentite



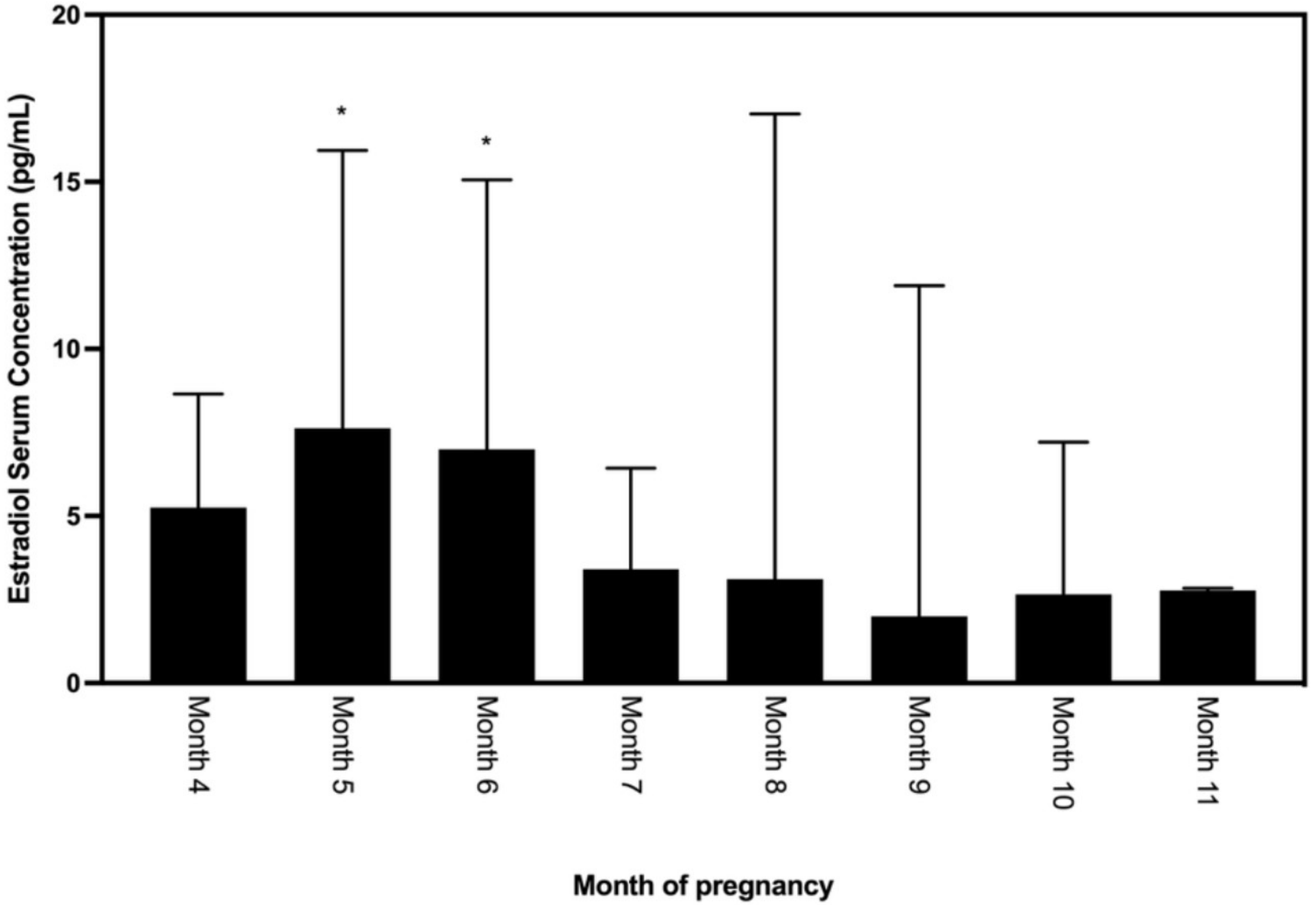
ESTRADIOL



ESTRONE

Résultats :

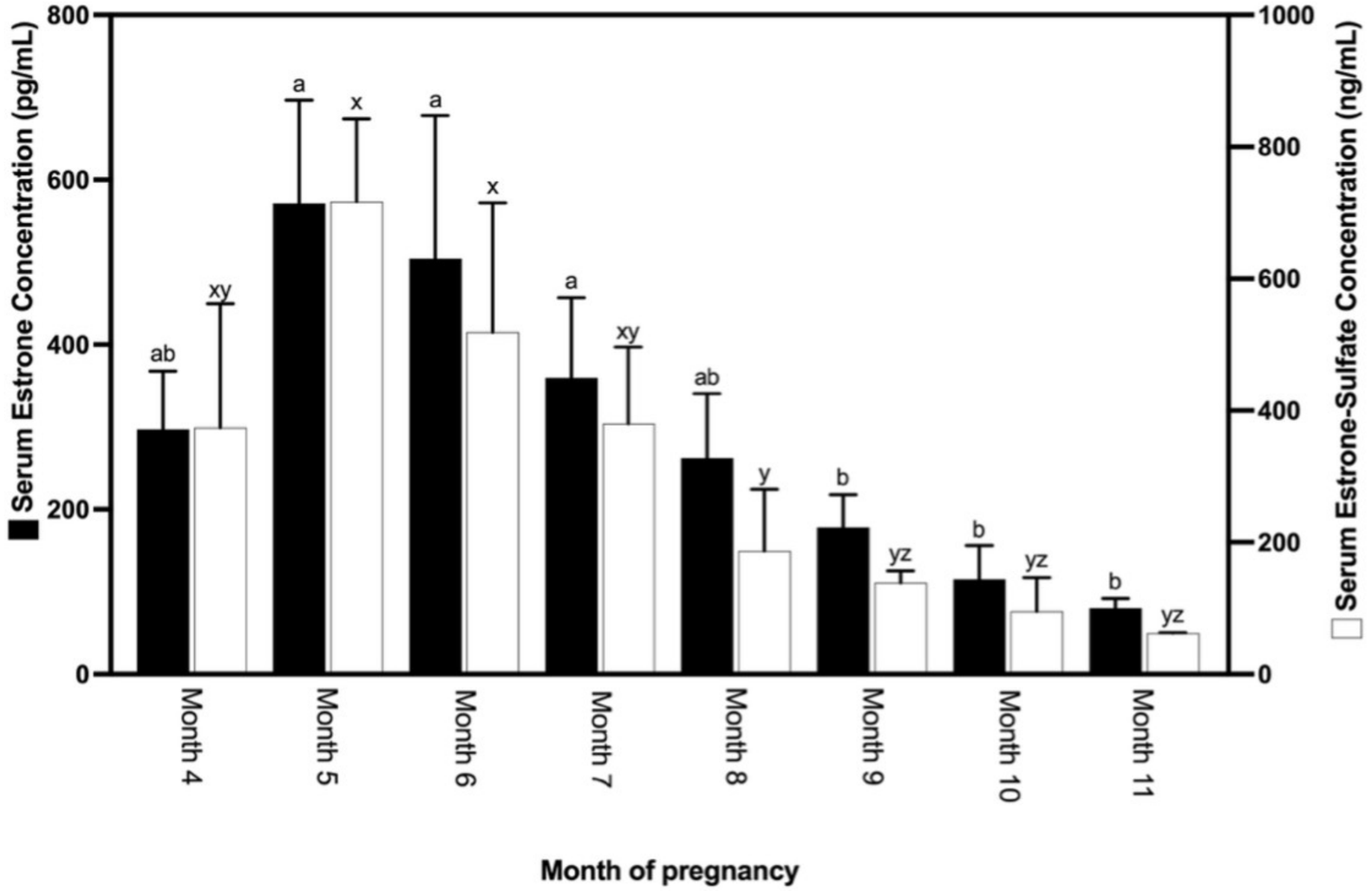
Evolution of E2 serum concentration from 4 to 11 months of pregnancy in



Results show median E2 concentration (pg/mL) and quartile ranges for each month of pregnancy.
E2 medians with the superscript * are significantly different from others ($p<0,05$).

Résultats :

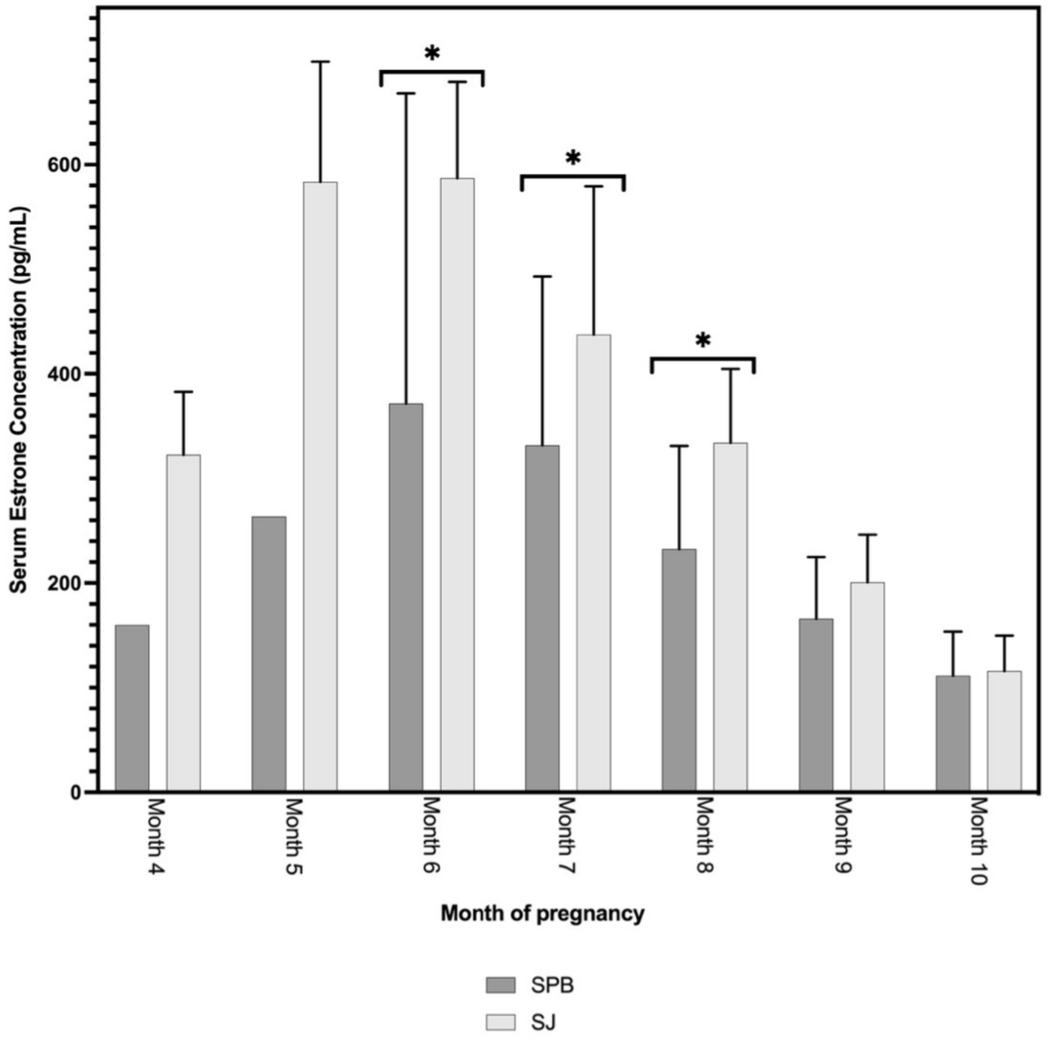
Evolution of E1 and E1S serum concentration from 4 to 11 months of pregnancy in mares



Results show median concentration of E1 (▪) in pg/mL and E1S (□) in ng/mL and quartile ranges for each month of pregnancy.
For E1, values with different superscript (a,b) statistically different (p. value<0,05).
For E1S, values with different superscript (x,y,z) statistically different (p. value <0,05).

Résultats :

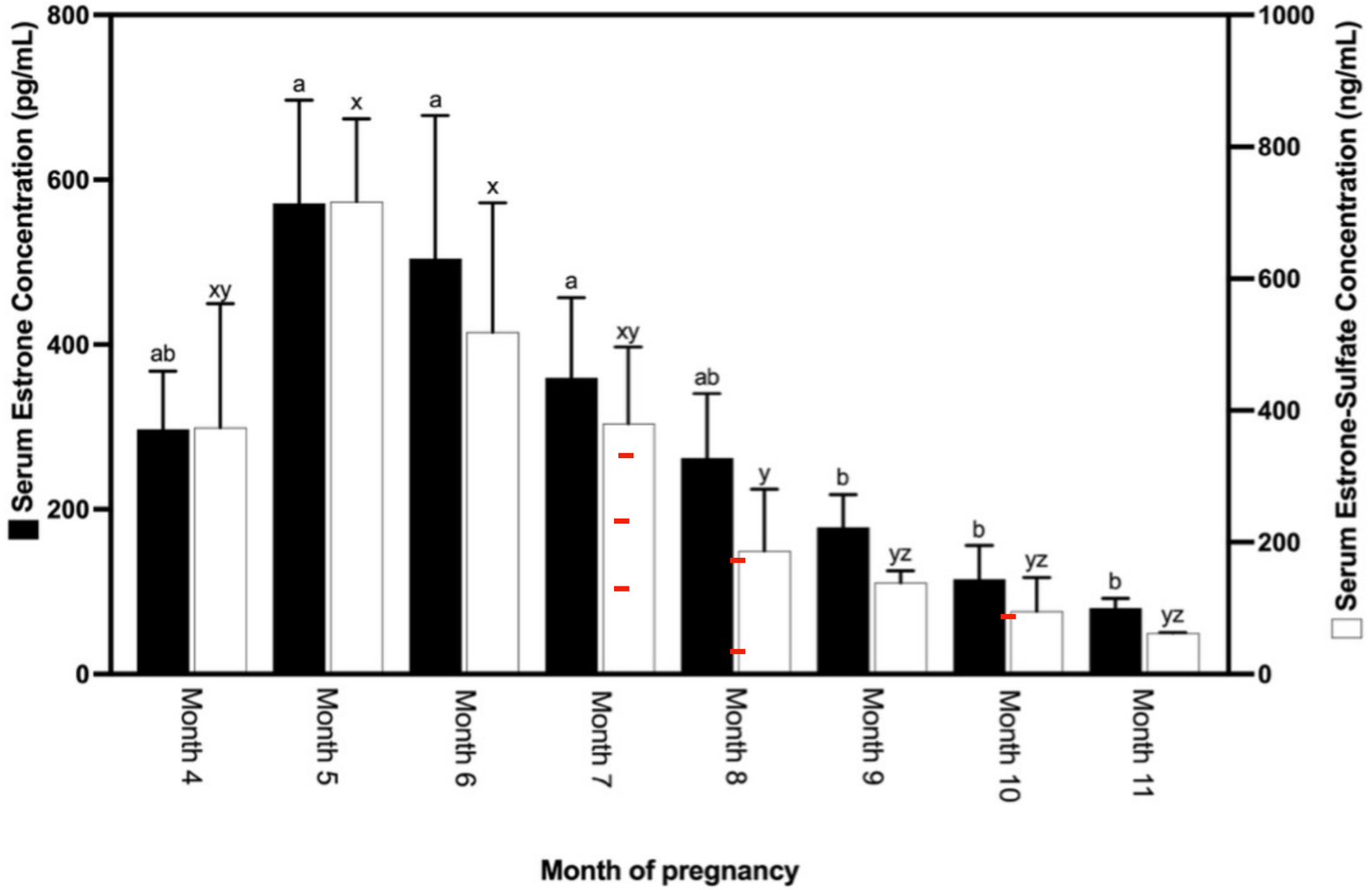
Comparison of E1 concentration between Spanish Pure-Breeds (SPB) and Show Jumping (SJ) mares at 4, 5, 6, 7, 8 and 10 months of pregnancy.



Results show median concentration of E1 (pg/mL) and quartile range for each month of pregnancy.
The superscript * indicates a statistical ($p<0,05$) difference in E1 medians between SPB (Spanish Pure-Breed) an SJ (Showjumping) mares at the same month of pregnancy.

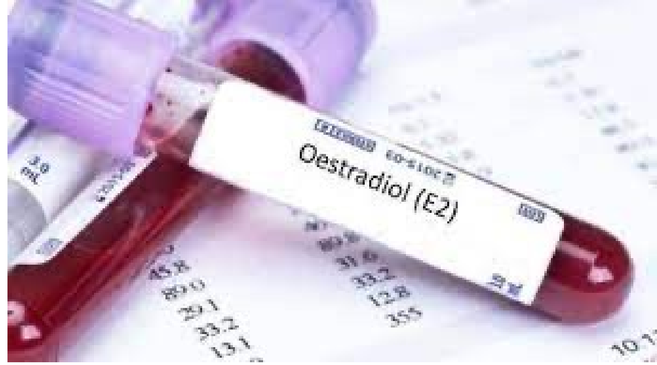
Résultats :

Evolution of E1 and E1S serum concentration from 4 to 11 months of pregnancy in mares



Results show median concentration of E1 (▪) in pg/mL and E1S (□) in ng/mL and quartile ranges for each month of pregnancy.
For E1, values with different superscript (a,b) statistically different ($p<0,05$).
For E1S, values with different superscript (x,y,z) statistically different ($p<0.05$).

Discussion :



La production d'E2 < à E1 = ↑hydroxystéroïde déshydrogénase 17B2 par l'endomètre → synthèse d'E1 > E2 *(Loux et al., 2020)*.

Pics à 5 mois de gestation, valeurs (ng/mL)

+/- = à celles de RIA ou ELISA.

Legacki *et al.* (2019), valeurs en µg/mL (LC-MS/MS) → Résultats significatif (n=6) ? Sensibilité ? Nutrition ? Phyto-œstrogènes ?

Effets sur les concentrations en œstrogènes :

E1 et E1S corrélées MAIS ↓↓↓ E1S

Expression limitée des gènes placentaires liés à la sulfonation après 6 mois de gestation *(Loux et al., 2020)*.

Effet race entre 6 et 8 mois de gestation sur les concentrations en œstrogènes mais pas sur E1S.

Sulfonation est un facteur limitant *(Loux et al., 2020)* et explique en partie la variabilité de l'E1 et l'E2.

Conclusion :

<u>Diagnostic hormonal de placentite :</u>

LC-MS/MS

Dosage de l'E2 : NON, valeurs = œstrus (RIA) *(Bergfelt et al., 2001 ; Ginther, 2017)*

Dosage de l'E1 : Oui mais effet race entre 6 et 8 mois

Dosage de l'E1S

Pas d'effet race

Valeurs >>> œstrus (RIA) *(Oxender et al., 1975)*

Placentite : ↓ E1, E2 et E1S (Canisso *et al.*, 2017) => résultats variables et n = 3 donc non significatif

Perspectives :

Screening des œstrogènes (non) sulfo-conjugués (LC-MS/MS) chez des juments saines

Effet race

Effet sexe

Effet parité

Effet âge

Screening des œstrogènes atypiques et de leur précurseurs androgéniques (HR-MS et LC-MS/MS)

Modèle de culture cellulaire d'allanto-chorion : étude des modifications de productions hormonales en cas d'application de différents stress

Références bibliographiques :

Albrecht, E. D., Robb, V. A., & Pepe, G. J. (2004). Regulation of placental vascular endothelial growth/permeability factor expression and angiogenesis by estrogen during early baboon pregnancy. The Journal of clinical endocrinology and metabolism, 89(11), 5803–5809. https://doi.org/10.1210/jc.2004-0479

Bailey, C.S., Heitzman, J.M., Buchanan, C.N., Bare, C.A., Sper, R.B., Borst, L.B., Macpherson, M., Archibald, K. and Whitacre, M. (2012). Diagnosis of equine placentitis using B-mode and Doppler ultrasonography. Equine Vet J, 44: 88-94. https://doi.org/10.1111/j.2042-3306.2012.00658.x

Barnes, M., Fite, C., Tibary, A. (2005). Trans-rectal ultrasonographic evaluation of the placenta in Arabian and pony mares in mid-to-late gestation. Theriogenology, 64.

Bergfelt, D.R., Adams, G.P. (2011). Pregnancy in Equine reproduction. 2nd éd. Wiley-Blackwell, 2065-2079.

Bucca, S., Fogarty, U., Collins, A., & Small, V. (2005). Assessment of feto-placental well-being in the mare from mid-gestation to term: transrectal and transabdominal ultrasonographic features. Theriogénology, 64(3), 542–557. https://doi.org/10.1016/j.theriogenology.2005.05.011

Brinsko, S., Blanchard, T., Dickson, V., Schumacher, J., Love, C., Hinrichs, K., Hartman, D., Manual of Equine Reproduction (Third Edition), 2011, ISBN 9780323064828, https://doi.org/10.1016/B978-0-323-06482-8.00003-X.

Bucca S. (2006). Diagnosis of the compromised equine pregnancy. The Veterinary clinics of North America. Equine practice, 22(3), 749–761. https://doi.org/10.1016/j.cveq.2006.07.006

Campos, I. S., de Souza, G. N., Pinna, A. E., & Ferreira, A. (2017). Transrectal ultrasonography for measuring of combined utero-placental thickness in pregnant Mangalarga Marchador mares. Theriogenology, 96, 142–144. https://doi.org/10.1016/j.theriogenology.2017.04.013

Canisso, I.F., Ball, B.A., Esteller-Vico, A., Williams, N.M., Squires, E.L. and Troedsson, M.H. (2017). Changes in maternal androgens and oestrogens in mares with experimentally-induced ascending placentitis. Equine Vet J, 49: 244-249. https://doi.org/10.1111/evj.12556

Chavatte-Palmer, P., Duchamp, G., Palmer, E., Ousey, J. C., Rossdale, P. D., & Lombès, M. (2000). Progesterone, oestrogen and glucocorticoid receptors in the uterus and mammary glands of mares from mid- to late gestation. Journal of reproduction and fertility. Supplement, (56), 661–672.

Colón, J. (2008). Trans-Rectal Ultrasonographic Appearance of Abnormal Combined Utero-Placental Thickness in Late-Term Gestation and its Incidence During Routine Survey in a Population of Thoroughbred Mares. Proc Am Assoc Equine Pract, 54:279-285.

Références bibliographiques :

Conley A. J. (2016). Review of the reproductive endocrinology of the pregnant and parturient mare. Theriogenology, 86(1), 355–365. https://doi.org/10.1016/j.theriogenology.2016.04.049

Conley, A. J., & Ball, B. A. (2019). Steroids in the establishment and maintenance of pregnancy and at parturition in the mare. Reproduction (Cambridge, England), 158(6), R197–R208. https://doi.org/10.1530/REP-19-0179

Coutinho da Silva, M. A., Canisso, I. F., MacPherson, M. L., Johnson, A. E., & Divers, T. J. (2013). Serum amyloid A concentration in healthy periparturient mares and mares with ascending placentitis. Equine veterinary journal, 45(5), 619–624. https://doi.org/10.1111/evj.12034

Cox, J.E. (1975). Oestrone and equilin in the plasma of the pregnant mare. Journal of Reproduction and fertility. Supplement, (23):463-468.

Dufour, P., Courtois, J., Seynaeve, Y., Peeters, S., Le Goff, C., Cavalier, E., & Ponthier, J. (2021). Development and validation of a liquid chromatography coupled to mass spectrometer (LC-MS) method for the simultaneous quantification of estrone-3-sulfate, progesterone, estrone and estradiol in serum of mares and American bisons. Research in veterinary science, 136, 343–350. Advance online publication. https://doi.org/10.1016/j.rvsc.2021.03.014

Esteller-Vico, A., Ball, B. A., Troedsson, M., & Squires, E. L. (2017). Endocrine changes, fetal growth, and uterine artery hemodynamics after chronic estrogen suppression during the last trimester of equine pregnancy. Biology of reproduction, 96(2), 414–423. https://doi.org/10.1095/biolreprod.116.140533

Fowden, A. L., Forhead, A. J., & Ousey, J. C. (2008). The Endocrinology of equine parturition. Experimental and clinical endocrinology & diabetes : official journal, German Society of Endocrinology [and] German Diabetes Association, 116(7), 393–403. https://doi.org/10.1055/s-2008-1042409

Kasman, L. H., Hughes, J. P., Stabenfeldt, G. H., Starr, M. D., & Lasley, B. L. (1988). Estrone sulfate concentrations as an indicator of fetal demise in horses. American journal of veterinary research, 49(2), 184–187.

Kimura, Y., Haneda, S., Aoki, T., Furuoka, H., Miki, W., Fukumoto, N., Matsui, M., & Nambo, Y. (2018). Combined thickness of the uterus and placenta and ultrasonographic examinations of uteroplacental tissues in normal pregnancy, placentitis, and abnormal parturitions in heavy draft horses. Journal of equine science, 29(1), 1–8. https://doi.org/10.1294/jes.29.1

LeBlanc M. M. (2010). Ascending placentitis in the mare: an update. Reproduction in domestic animals = Zuchthygiene, 45 Suppl 2, 28–34. https://doi.org/10.1111/j.1439-0531.2010.01633.x

Legacki, E. L., Corbin, C. J., Ball, B. A., Wynn, M., Loux, S., Stanley, S. D., & Conley, A. J. (2016). Progestin withdrawal at parturition in the mare. Reproduction (Cambridge, England), 152(4), 323–331. https://doi.org/10.1530/REP-16-0227

Legacki, E. L., Scholtz, E. L., Ball, B. A., Stanley, S. D., Berger, T., & Conley, A. J. (2016). The dynamic steroid landscape of equine pregnancy mapped by mass spectrometry. Reproduction (Cambridge, England), 151(4), 421–430. https://doi.org/10.1530/REP-15-0547

Références bibliographiques :

Legacki, E. L., Scholtz, E. L., Ball, B. A., Esteller-Vico, A., Stanley, S. D., & Conley, A. J. (2019). Concentrations of sulphated estrone, estradiol and dehydroepiandrosterone measured by mass spectrometry in pregnant mares. *Equine veterinary journal*, *51*(6), 802–808. https://doi.org/10.1111/evj.13109

Morris, S., Kelleman, A. A., Stawicki, R. J., Hansen, P. J., Sheerin, P. C., Sheerin, B. R., Paccamonti, D. L., & LeBlanc, M. M. (2007). Transrectal ultrasonography and plasma progestin profiles identifies feto-placental compromise in mares with experimentally induced placentitis. *Theriogenology*, *67*(4), 681–691. https://doi.org/10.1016/j.theriogenology.2006.05.021

Murase, H., Niwa, H., Katayama, Y., Sato, F., Hada, T., & Nambo, Y. (2015). A clinical case of equine fungal placentitis with reference to hormone profiles and ultrasonography. *Journal of equine science*, *26*(4), 129–133. https://doi.org/10.1294/jes.26.129

Niklaus, A. L., Aberdeen, G. W., Babischkin, J. S., Pepe, G. J., & Albrecht, E. D. (2003). Effect of estrogen on vascular endothelial growth/permeability factor expression by glandular epithelial and stromal cells in the baboon endometrium. *Biology of reproduction*, *68*(6), 1997–2004. https://doi.org/10.1095/biolreprod.102.011288

Noakes, D. E., In Parkinson, T. J., & In England, G. C. W. (2019). Veterinary reproduction and obstetrics.

Ousey J. C. (2004). Peripartal endocrinology in the mare and foetus. *Reproduction in domestic animals* = *Zuchthygiene*, *39*(4), 222–231. https://doi.org/10.1111/j.1439-0531.2004.00507.x

Pashen, R. L., & Allen, W. R. (1979). The role of the fetal gonads and placenta in steroid production, maintenance of pregnancy and parturition in the mare. *Journal of reproduction and fertility. Supplement*, (27), 499–509.

Pashen, R. L., & Allen, W. R. (1979). Endocrine changes after fetal gonadectomy and during normal and induced parturition in the mare. *Anim Reprod Sci*, 2:271–288.

Peugnet P. Origines développementales des anomalies de l'homéostasie glucidique, de la croissance osseuse et prédisposition à l'ostéochondrose chez le poulain. Université Paris Sud - Paris XI; Université de Liège. 2014.

Références bibliographiques :

Raeside J. I. (2017). A Brief Account of the Discovery of the Fetal/Placental Unit for Estrogen Production in Equine and Human Pregnancies: Relation to Human Medicine. *The Yale journal of biology and medicine*, *90*(3), 449–461.

Renaudin, C. D., Troedsson, M. H., Gillis, C. L., King, V. L., & Bodena, A. (1997). Ultrasonographic evaluation of the equine placenta by transrectal and transabdominal approach in the normal pregnant mare. *Theriogenology*, *47*(2), 559–573. https://doi.org/10.1016/s0093-691x(97)00014-9

Robb, V. A., Pepe, G. J., & Albrecht, E. D. (2004). Acute temporal regulation of placental vascular endothelial growth/permeability factor expression in baboons by estrogen. *Biology of reproduction*, *71*(5), 1694–1698. https://doi.org/10.1095/biolreprod.104.030882

Robb, V. A., Pepe, G. J., & Albrecht, E. D. (2007). Placental villous vascular endothelial growth factor expression and vascularization after estrogen suppression during the last two-thirds of baboon pregnancy. *Endocrine*, *31*(3), 260–267. https://doi.org/10.1007/s12020-007-0036-5

Santschi, E. M., LeBlanc, M. M., & Weston, P. G. (1991). Progestagen, oestrone sulphate and cortisol concentrations in pregnant mares during medical and surgical disease. *Journal of reproduction and fertility. Supplement*, *44*, 627–634.

Satué, K., Domingo, R., & Redondo, J. I. (2011). Relationship between progesterone, oestrone sulphate and cortisol and the components of renin angiotensin aldosterone system in Spanish purebred broodmares during pregnancy. *Theriogenology*, *76*(8), 1404–1415. https://doi.org/10.1016/j.theriogenology.2011.06.009

Senger PL. Pathways to pregnancy and parturition. 3rd ed. 2015.

Shikichi, M., Iwata, K., Ito, K., Miyakoshi, D., Murase, H., Sato, F., Korosue, K., Nagata, S., & Nambo, Y. (2017). Abnormal pregnancies associated with deviation in progestin and estrogen profiles in late pregnant mares: A diagnostic aid. *Theriogenology*, *98*, 75–81. https://doi.org/10.1016/j.theriogenology.2017.04.024

Souza, A.M., Winter, G.H.Z., Garbade, P., Wolf, C.A., Jobim, M.I.M., Gregory, R.M., Mattos, R.C. 2010. Ultrasonography evaluation of the Criollo mare placenta. Anim Reprod Sci, 121:S320-321.

Terqui, M., & Palmer, E. (1979). Oestrogen pattern during early pregnancy in the mare. *Journal of reproduction and fertility. Supplement*, (27), 441–446