Early castration in foals: consequences on physical and behavioural development

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Abstract

Background: The impact of very early castration of foals has not yet been studied despite

the many positive effects observed in dogs and cats.

Objectives: To compare castration at 3 days and 18 months and assess their subsequent

morphological and behavioural development.

Study design: Randomised, blinded clinical study.

Methods: Twenty-two Welsh ponies underwent either early (3 days old, EC group, n=11) or

traditional (18 months old, TC group, n=11) castration. Animals were followed up to three

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years of age. All ponies were castrated using a primary closure technique under general anaesthesia. Weight and morphometric measurements were monitored monthly from birth until 8 months of age in both groups. Then, measurements were taken every 3 months until 2 years of age and then every 6 months until 3 years of age. Temperament tests were performed on all animals when they were 1 and 3 years old.

Results: No differences were observed between the EC and TC groups in terms of physical development from birth until 40 months of age, or in terms of temperament and behaviour at either 1 or 3 years of age.

Main limitations: The study included only one breed (Welsh ponies) and only 22 animals that were castrated before 2 years of age, precluding comparison with castration performed at older ages.

Conclusions: We demonstrate that early castration at three days does not interfere with morphological or behavioural development.

Introduction

The castration of horses has been performed since antiquity aiming to obtain calmer animals that are more attentive during sporting, agricultural or leisure activities and therefore less dangerous. It also facilitates the constitution of herds without the risk of undesirable mating.

Castration is one of the most common surgeries in equine veterinary practice. Despite this, complications are not uncommon, with potential for fatal outcomes and/or complaints against practitioners.^{1,2} Castration may be delayed until the age of 2 to 3 years to obtain stallion-like morphological characteristics but is generally recommended before the age of 2 years.² The anticipated use of the horse can influence decision-making, especially in racing where successful colts can go on to a breeding career. According to Line et al.,³ male horses

not intended for use in breeding are castrated before puberty to prevent the development of their libido and the more aggressive behaviour of a stallion. However, in their study, they concluded that there was no behavioural difference between animals castrated before 2 years and at 3 years of age.³

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The prepubertal gonadectomy of dogs and cats began in the mid-1980s, and numerous articles have shown the many benefits and few side effects of these early-age interventions.^{4,5} In equine primary care practice, anecdotal reports suggest some veterinarians are castrating male foals starting at the age of 6 months however, there are no studies on early castration in horses and the equine sector has no evidence-based recommendations. There are open questions regarding the effects on dam-foal relations (with possible foal rejection after the intervention) and the subsequent behaviour and physiology of the foal. Early surgery, although a convenient intervention, can also be rejected by veterinarians and owners due to fear of perioperative complications. The objective of our study was to evaluate the influence of early-age castration (at 3 days of age) compared to traditional castration (at 18 months of age) with regard to morphological and behavioural development.

Materials and Methods

Animals

This study used 22 male Welsh ponies from an experimental herd and included foals born in three consecutive seasons. Pregnant mares were kept together and foaled in the field. Only when care was needed for a neonate were the dam and foal kept indoors for two or three days and fed hay and pellets. Water was provided ad libitum. The colts were gradually weaned at 6 months ⁶ and weaning involved progressive habituation to separation using a fence line during the month preceding definitive separation. After weaning, animals were placed in a

yearling group, and then 2- and 3-year-old ponies were housed together. Horses were kept at pasture from March to November, with hay and water provided *ad libitum* and were housed in stalls with outside access from November to March. Ponies underwent either early (3 days or neonates, EC group, n = 11) or traditional (18 months or yearling, TC group, n = 11) castration and were followed up to the age of 3 years to compare castration as commonly performed in practice with neonatal foal castration. The scrotal position of the testicles, light bodyweight (bwt) of the animals and subsequent expected ease of the procedure accounted for the choice of three days of age to compare with a more conventional age group. The testicles were being examined histologically as part of another study comparing development between the two groups.

Castration protocol and peri-operative management

<u>Anaesthesia</u>

From birth foals were accustomed to the human voice and presence by daily visual, vocal and physical contacts. Before any manipulation of the neonates, the mother was sedated with intravenous (I.V.) romifidine (Sedivet, 0.035 mg/kg bwt, Boehringer Ingelheim).

The yearlings were left in their social group until they were fasted for 12 hours overnight and then premedicated. In the days before the operation, they were examined clinically and weighed.

After a catheter was placed in the jugular vein and warm saline infusion (sodium chloride solution 0.9%, 5 ml/min, Virbac) was started, the following I.V. injections were given at five-minute intervals: i) diazepam for neonates (Valium, 0.08 mg/kg bwt, Laboratoire Roche) for sedation and muscular relaxation (in the case of excessive foal agitation, the dose of Valium was increased);⁷ ii) romifidine for yearlings (Sedivet, 0.05 mg/kg bwt, Boehringer Ingelheim)

for sedation; iii) butorphanol (Dolorex, 0.015 mg/kg bwt for neonates, 0.02 mg/kg bwt for yearlings, Intervet) for analgesia⁸ (this injection was repeated immediately after the operation); ⁹ and iv) ketamine (Imalgene 1000, 3-5 mg/kg bwt for neonates, 2.2 mg/kg bwt for yearlings, Merial SAS) for the induction of anaesthesia.¹⁰

Analgesia at the operating site was achieved with an injection of local anaesthetics (lidocaine, Lurocaine, 5 ml/neonate, 10 ml/yearling, Vetoquinol) into the subcutaneous tissue and around the cord after removal of the testicles. The gonads were not infiltrated with lidocaine prior to excision to avoid impact on subsequent histological examination. Furosemide (Dimazon, 1 mg/kg bwt, Intervet) was given intravenously, at the end of procedure to control postsurgical oedema and was administered again in the following days if necessary.

After the animal regained consciousness, a nonsteroidal anti-inflammatory drug (flunixin meglumine, Finadyne, 1 mg/kg bwt, Intervet) was administered intravenously for analgesia.¹¹ This injection was repeated on subsequent days if signs of pain persisted. In the case of excessive agitation before surgery, the dose of romifidine and/or butorphanol was increased; this occurred in three neonates and five yearlings. During the operation, based on clinical observations (movement, respiratory rate, cranial nerve reflexes, movement after surgical stimulation, increased tension on the leg rope), incremental I.V. doses of ketamine were administered. No differences in anaesthetic or analgesic requirements were observed between the groups.

Surgery

All animals were castrated using a primary closure technique under general anaesthesia. Two experienced veterinary surgeons were involved in this project. Twelve to four hours before surgery, an intramuscular injection of antibiotic was administered (Depocilline, benzylpenicillin, 10 mg/kg bwt, Intervet). For yearlings, antibiotic therapy was continued for two days following surgery and for one to four days in both groups in the case of bleeding or risk of contamination. The horses were positioned in dorsal recumbency. The eyes were covered to limit sensory stimulation. During surgery, while keeping a distance and maintaining appropriate asepsis, visual and olfactory contact between the dam and her foal was never broken to preserve the dam-foal bond and prevent rejection of the foal after surgery.

The scrotal and inguinal areas were surgically prepared and draped. Briefly, the testicles were pushed cranially, and a 4- to 5-cm incision was made through the prescrotal skin and underlying subcutaneous tissues, which could be further dissected bluntly using Metzenbaum scissors. One testicle was then exteriorised under gentle pressure, and the parietal tunica vaginalis was incised to expose the testicle and epididymis. The vascular cord was clamped and ligated separately from the cremaster muscle. After testicular removal, the parietal tunica vaginalis was sutured, and the second testicle was dissected in the same manner through the same skin incision. After the injection of local anaesthetic in the dead space, the subcutaneous tissue was closed with a continuous suture, and finally, the skin was closed with three or four sutures. All ligations were performed with Surgicryl (polyglycolic acid, SMI) USP 0 in neonates and Surgicryl USP 2 in yearlings.

Neonates were returned to their dam as soon as the first signs of recovery were observed and were allowed to suckle as soon as they were able to stand. They were kept in a stall for 24 hours before returning to the pasture if no signs of pain or discomfort appeared.

Yearlings were kept in their stall for 24 hours. Then they were placed with another castrated individual for ten days in spacious stalls with paddock access before joining their social group in the pasture or stall. Water was available as soon as effective recovery was observed, and

hay was gradually reintroduced within four hours of the intervention. Wounds examination and monitoring for signs of complications were performed twice a day for the next five days.

Data collection

Morphometric measurements

Weight and morphometric monitoring were performed monthly from birth to 8 months of age in both groups. Then, measurements were taken every 3 months until 2 years of age and then every 6 months until 3 years of age.

The variables measured were as follows: bwt (kg), measured using an electronic livestock scale; height at the withers (cm); body length (cm), measured from the greater tubercle of the humerus to the point of the ischial tuberosity; chest perimeter (cm), measured through the webbing passage and behind the withers; chest width (cm), measured as the distance between the right and left greater tubercle of the humerus; hip width (cm), measured as the distance between the right and left coxal tuberosity of the ilium; distal limb length (cm), measured as the distance from the ground to the olecranon tuberosity; and metacarpal width (cannon bone width, cm), measured with callipers.

Temperament tests

Tests were performed at 1 year of age to ensure that there were no side effects of the paediatric surgery on temperament and at 3 years of age, the age of breaking-in, to evaluate whether the age of castration could alter the further use of the animals by humans.

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The temperament traits assessed were reactivity to humans, sensory sensitivity, gregariousness, fearfulness towards novelty and suddenness, and activity. These tests have been previously fully described¹²⁻¹⁵ and are specified in Supplementary Item 1.

Briefly, reactivity to humans was measured based on the number of contacts with a passive human during a 180 s period. Sensory sensitivity was evaluated as the number of cutaneous reactions to von Frey filaments applied at the withers. Gregariousness was assessed by the number of neighs during a 90 s period of isolation. Fearfulness towards novelty and suddenness was assessed by monitoring the following four measurements: number of looks at a novel object during a period of 180 s; number of contacts with a novel object during a period of 180 s; number of contacts with a novel object during a period of 180 s; number of contacts with a novel object during a period of 180 s; time interval required to cross a novel area (seconds); and startle response intensity in a suddenness test (notation of reactions according to their violence, from no reaction to a very violent half-turn or even a fall). Activity was measured as the number of moves in six delimited areas on the ground surface during habituation, passive human testing, novel object testing and social isolation.

Data analysis:

Temperament data were compared between the two groups using the nonparametric Wilcoxon-Mann-Whitney exact test for two independent groups with SAS[®] software (SAS Institute, Inc.) and the npar1way procedure (Wilcoxon option).

The evolution of foal bwt (from birth) and morphometric measurements (from 1 week of age) to 40 months of age were compared between the two groups using a nonparametric analysis of longitudinal data in a factorial experiment with small samples using SAS[®] software (SAS Institute, Inc.) and the F1 LD F1 macro (version 1.1, written by Steffen Ballerstedt, January

1998, last version by Carola Werner, February 2005, Department of Medical Statistics, University of Gottingen). The effects of group (EC or TC), animal age (time effect) and interactions between group and age were included in the model.

Results

Surgeries

We did not observe any cases of prolonged hypothermia following anaesthesia. All foals happily returned to their mother after surgery, and no cases of rejection were observed. To facilitate the recovery process, each yearling was able to hear his companion the first 24 hours after surgery. We did not observe any sweating, colic or behavioural problems suggestive of pain after the surgery in neonates or yearlings. Apart from a few cases of slight oedema in 7 yearlings and 2 neonates, which quickly resolved, no wound-related complications were observed.

Temperament tests and behaviour

Descriptive statistics (mean, median, minimum, maximum, Q1, Q3) for the results of temperament tests performed when the horses were one or three years old, are presented in Tables 1-2 and in Figures S1-S5 (boxplots). No differences in temperament were observed between the two groups, regardless of the measurement (reactivity to humans, sensory sensitivity, fearfulness-reaction to novelty and to suddenness, gregariousness, activity), at either one year (Table 1) or three years (Table 2) of age.

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Descriptive statistics (mean, median, minimum, maximum, Q1, Q3) for results of morphometric measurements, performed at birth (bwt) or 1 week to 40 months of age, are presented in Figures 1-2 (boxplots) and Table S1. No differences were observed between the two groups in either bwt at birth to 40 months of age (Figure 1) or other morphometric measurements from 1 week to 40 months of age (Figure 2), except for the hip width, with an interaction between group and age (p=0.04, Figure 2f).

Discussion

An open castration technique with a prescrotal approach and primary closure under general anaesthesia was applied to both groups. This technique was selected due to the lower rate of associated complications and quicker recovery.¹⁶ In addition, using the same technique in both groups allowed an independent assessment of complications.

Preanaesthetic stress should be avoided as much as possible as it may interfere with the quality of narcosis, particularly in neonates. Therefore, transposing the general guidelines in young dogs and cats,⁵ foals were housed with their dam in a quiet location, and manipulations were limited as much as possible before anaesthesia induction. As they are still in a period where their physiology and metabolism are adjusting to their new extrauterine environment, neonates are prone to hypothermia and hypoglycaemia during anaesthesia, particularly under field conditions. Considering the unique metabolism and excretion processes of foals at a very early age, anaesthetic doses were kept as minimal as possible. Regarding hypothermia, surgeries were performed in spring, outside but in a place sheltered from the wind, and the foals were placed on a table covered with blankets for insulation. The rectal temperature was monitored before and after surgery. Because glycogen depletion should be avoided and as

regurgitation and intestinal tympany are uncommon during anaesthesia in foals without preexisting gastrointestinal obstruction,¹⁷ preanaesthetic fasting is no longer recommended, but caution should be used when induction occurs soon after the animal has nursed.¹⁸ Under our conditions, foals were prevented from nursing during the preparation time and were allowed to suckle as soon as they recovered from anaesthesia. Although it was not specifically measured, as all foals returned to suckling very shortly after recovery, no general difference in their behaviour was noticed by comparison with untreated foal, and no signs of foal rejection by the mare were observed, it can be considered that early castration did not negatively affect the dam-foal bond. This is somewhat supported by the similar growth of foals in both groups, which confirms that the treated foals nursed normally after surgery.

No pre or intraoperative complications were observed, and all the foals and yearlings recovered very quickly. However, we could have optimised the analgesia by injecting lidocaine or mepivacaine into the testicle, which could easily be implemented in field practice to improve the animal's comfort. ^{19,20} As in previous reports,² we observed no long-term complications, such as oedema, weight loss, castration adhesions or infections at the surgical site, in our study. Castration of animals at 2 to 3 years of age has been shown to have an increased risk of eventration in draught horses.¹ We speculate that very early castration with a primary closure technique may be particularly beneficial in these breeds and should be evaluated against later castration.

Our observations are based on only twenty-two animals born in one single herd of Welsh ponies. While having one single breed represented is an obvious limitation, the common housing, breeding and diet conditions among all animals enrolled are strengths. With more than 20 years of experience each, both participating surgeons agreed that they subjectively felt that early castration was easier and smoother than late castration. While this might have been highlighted by a shorter surgical time, inconsistent recording of this parameter prevents statistical confirmation of this subjective impression.

No mare rejected her foal after the castration surgery, and no foal showed any behavioural problems after recovery from general anaesthesia (refusal to suckle, hyperactivity, prostration, disordered movements, etc.). Regarding the temperament tests, the variables recorded are considered good indicators of reactivity to humans, sensory sensitivity, gregariousness, fearfulness and activity, due to their stability across situations and over time.¹²⁻¹⁵ Early castration therefore had no significant influence on behavioural development.

The selected mares were multiparous to avoid differences in birth weight and the subsequent development of the foals.²¹ No differences were observed in terms of physical or behavioural development in the EC group compared to the TC group during the morphometric evaluation or temperament tests carried out at 1 and 3 years of age.

It is important to note that this early castration was performed on animals that were closely monitored by professionals. Further studies are needed to confirm the feasibility in routine field practice. Owners may be reluctant to make the decision to operate on such young animals. This technique is therefore primarily intended for professionals.

These initial results relate only to morphometry and weight but do not prejudge bone development. Further studies are needed to confirm the absence of an effect on bone development and metabolism.

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Conclusion

Performing castration at a younger age was safe in healthy neonates and was technically easier than in yearlings. There were no significant differences identified in morphological or behavioural development between the groups.

Authors' declaration of interests

No competing interests have been declared.

Ethical animal research

All experimental procedures were approved by the Val de Loire Ethics Committee (authorization N° APAFIS#4530-2016031415011475 v3).

Informed consent

Not applicable.

Data availability statement

The data that support the findings of this study are openly available in data.inrae.fr at https://data.inrae.fr/dataset.xhtml?persistentId=doi:10.15454/9C3GHF

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Authorship

J. Cognie contributed to the study design, study execution, surgeries, preparation of the manuscript and final approval. S. Freret contributed to the preparation of the manuscript, data analysis and interpretation. L. Lansade contributed to the study design with regard to

the temperament tests and data analysis and to the preparation of the manuscript. C. Parias contributed to the study design with regard to the temperament tests. P. Barriere, A. Gesbert and F. Reigner contributed to the study design, study execution and preparation of the manuscript. S. Deleuze contributed to the study design, surgeries, writing and final approval of the manuscript.

Tables

Table 1: Descriptive statistics and comparison of temperament data, between the EC (n=11) and TC (n=11) groups at one year of age

(Wilcoxon-Mann-Whitney test, exact, bilateral)

		One year old (comparison with Wilcoxon–Mann–Whitney test, exact, bilateral)												
		EC group	(n=11)	TC group										
		Mean	Median	Min	Max	Q1	Q3	Mean	Median	Min	Max	Q1	Q3	p value
Reactivity to human	Number of contacts with a passive human	18.09	15	0	57	6	29	7.91	8	1	21	2	12	p=0.1
	Time interval to touch the horse (head)	12.36	6	4	36	4	19	8	6	4	34	4	6	p=0.5
Sensory sensitivity	Intensity of reaction to von Frey Filaments	1.64	2	0	3	1	3	1.27	1	0	3	0	2	p=0.6
Fearfulness- reaction to novelty	Number of looks at a novel object	6.45	6	0	15	1	10	4.55	3	0	17	2	7	p=0.5
	Number of contacts with a novel object	5.55	2	0	33	0	8	7.09	0	0	25	0	14	p=0.8
	Time interval to cross a novel area (seconds)	40.91	23	6	122	16	58	32.91	14	9	180	11	33	p=0.4
Fearfulness- reaction to suddenness	Startle response in a suddenness test	1.28	1	0.03	2.5	1	2	1.21	1	0.03	2	1	2	p=0.9
Gregariousness	Number of neighs during isolation	2.36	2	0	7	0	4	2.36	2	0	6	1	4	p=0.9
Activity	Activity (total number of movements)	72.09	72	45	108	52	83	77	80	18	152	43	98	p=0.9

Table 2: Descriptive statistics and comparison of temperament data, between the EC (n=11) and TC (n=11) groups at three years of age (Wilcoxon–Mann–Whitney test, exact, bilateral)

		Three years old (comparison with Wilcoxon–Mann–Whitney test, exact, bilateral)												
	EC group (n=11) TC group (n=11)													
		Mean	Median	Min	Max	Q1	Q3	Mean	Median	Min	Max	Q1	Q3	p-value
Reactivity to human	Number of contacts with a passive human	15.36	16	5	22	12	20	12.09	12	2	24	3	18	p=0.3
	Time interval to touch the horse (head)	7.36	8	4	11	6	8	11.45	9	5	30	6	16	p=0.1
Sensory sensitivity	Intensity of reaction to von Frey Filaments	1.82	2	0	4	1	3	1.36	1	0	3	1	2	p=0.4
Fearfulness- reaction to novelty	Number of looks at a novel object	7.91	8	1	14	5	12	8.27	9	2	14	4	11	p=0.8
	Number of contacts with a novel object	3.82	4	0	10	3	5	5.27	3	0	19	0	8	p=0.7
	Time interval to cross a novel area (seconds)	41.82	25	6	180	13	46	29.09	13	4	180	7	22	p=0.2
Fearfulness- reaction to suddenness	Startle response in a suddenness test	1.16	1	0.03	2.5	1	2	1.09	1	0.5	2	1	2	p=0.7
Gregariousness	Number of neighs during isolation	2	2	0	6	0	3	2.45	2	0	6	1	3	p=0.6
Activity	Activity (total number of movements)	80.45	78	46	112	72	96	70.82	68	51	117	55	77	p=0.1

Figure legends

Figure 1: Evolution of bodyweight from birth (month=0) to 40 months of age in the EC (n=11) and TC (n=11) groups (Black line = median, black circle = mean, nonparametric analysis of longitudinal data in a factorial experiment, testing the group effect, the age effect and the interaction between group and age).

Figure 2: Morphometric measures from 1 week (month=0.2) to 40 months of age in the EC (n=11) and TC (n=11) groups: (a) height at the withers (cm); (b) chest perimeter (cm); (c) body length (cm); (d) foreleg length (cm); (e) shoulder width (cm); (f) hip width (cm); and (g) cannon bone width (cm) (Black line = median, black circle = mean, nonparametric analysis of longitudinal data in a factorial experiment, testing the group effect, the age effect and the interaction between group and age).

Supporting Information

Supplementary Item 1: Description of the temperament tests.

Figure S1: Reactivity to humans (number of contacts with a passive human during a period of 180 s) in the EC (n=11) and TC (n=11) groups.

Figure S2: Fearfulness-reaction to novelty or suddenness in the EC (n=11) and TC (n=11) groups: (a) number of looks at a novel object during a period of 180 s; (b) number of contacts with a novel object during a period of 180 s; (c) time interval to cross a novel area (seconds); and (d) intensity of startle response in a suddenness test (0 = no reaction, 0.025 = head raise, 0.25 = head raise and startle, 0.5 = step back, 1 = quarter turn, 1.5 = violent quarter or half turn, 2 = violent half turn, 2.5 = very violent half turn, 3 = falls).

Figure S3: Sensory sensitivity (intensity of reaction = number of cutaneous reactions to the von Frey filaments) in the EC (n=11) and TC (n=11) groups.

Figure S4: Activity (number of areas crossed during habituation, passive human test, novel object test and social isolation) in the EC (n=11) and TC (n=11) groups.

Figure S5: Gregariousness (number of neighs during a 90 s period of isolation) in the EC (n=11) and TC (n=11) groups.

Table S1: Descriptive statistics for morphometric measurements in the EC (n=11) and TC (n=11) groups from birth (bodyweight) or the first week of life to 40 months of age.

- 1. Kilcoyne I. Equine castration: A review of techniques, complications and their management. Equine Vet. Educ. 2013; 25:476-482.
- 2. Moll HD, Pelzer KD, Pleasant RS, Modransky PD, May KA. A survey of equine castration complications. J. Equine Vet. Sci. 1995; 15:522-526.
- Line SW, Hart BL, Sanders L. Effect of prepubertal versus postpubertal castration on sexual and aggressive behaviour in male horses. J. Am. Vet. Med. Assoc. 1985; 186(3): 249-51.
- Howe LM, Slater MR, Boothe HW, Hobson HP, Holcom JL. Long-term outcome of gonadectomy performed at an early age or traditional age in dogs. J. Am. Vet. Med. Assoc. 2001; 218:217-221.
- 5. Root Kustritz MV. Early spay-neuter in the dog and cat. Veterinary Clinics of North America: Small Animal Practice 1999; 29:935-943.
- Lansade L, Foury A, Reigner F, Vidament M, Guettier E, Bouvet G, et al. Progressive habituation to separation alleviates the negative effects of weaning in the mother and foal. Psychoneuroendocrinology 2018; 97, 59-68. doi: 10.1016/j.psyneuen.2018.07.005.
- 7. De Vries AM, Thomson S, Taylor PM. Comparison of midazolam and diazepam as coinduction agents with ketamine for anaesthesia in sedated ponies undergoing field castration. Vet. Anaesth. Analg. 2015; 42:512-517.
- 8. Corletto F, Raisis AA, Brearley JC. Comparison of morphine and butorphanol as preanaesthetic agents in combination with romifidine for field castration in ponies. Vet Anaesth Analg 2005; 32:16-22.

- 9. Love EJ, Taylor PM, Clark C. Analgesic effect of butorphanol in ponies following castration. Equine Vet. J. 2009; 41(6):552-556.
- Hardardottir H, Blissitt K, Murison P, Olason S, Clutton R. A comparison of two ketamine doses for field anaesthesia in horses undergoing castration. Equine Vet. J. 2018; 51:458-463.
- Gobbi FP, Di Filippo PA, Mello LD, Lemos GB, Martins CB, Albernaz AP, Quirino CR.
 Effects of flunixin meglumine, firocoxib, and meloxicam in equines after castration. J.
 Equine Vet. Sci. 2020; 94, Article Number: 103204.
- 12. Lansade L, Bouissou M-F. Reactivity to humans: A temperament trait of horses which is stable across time and situations. Appl. Anim. Behav. Sci. 2008; 114:492.
- 13. Lansade L, Bouissou M-F, Erhard HW. Fearfulness in horses: A temperament trait stable across time and situations. Appl. Anim. Behav. Sci. 2008; 115:182.
- 14. Lansade L, Bouissou M-F, Erhard HW. Reactivity to isolation and association with conspecifics: A temperament trait stable across time and situations. Appl. Anim. Behav. Sci. 2008; 109:355.
- 15. Lansade L, Pichard G, Leconte M. Sensory sensitivities: Components of a horse's temperament dimension. Appl. Anim. Behav. Sci. 2008; 114:534.
- Crosa AT, Desjardins MR. Minimally invasive, compartmentalized, modified open castration technique with primary closure in equids. J. Am. Vet. Med. Assoc. 2018; 253 (7): 897-906.
- 17. Klein L. Anesthesia for neonatal foals. Vet Clin North Am Equine Pract, 1985. 1(1): p.77-89.

- 18. Bidwell LA, Bramlage LR, Rood WA. Equine perioperative fatalities associated with general anaesthesia at a private practice--a retrospective case series. Vet Anaesth Analg, 2007. **34**(1):23-30.
- Haga HA, Lykkjen S, Revold T, Ranheim B. Effect of intratesticular injection of lidocaine on cardiovascular responses to castration in isoflurane-anesthetized stallions. Am. J. Vet. Res. 2006; 67:403-408.
- 20. Crandall A, Hopster K, Grove A et al. Intratesticular mepivacaine versus lidocaine in anaesthetised horses undergoing Henderson castration. Equine Vet. J. 2020;**52**:805-810.
- 21. Robles M, Dubois C, Gautier C, Dahirel M, Guenon I, Bouraima-Lelong H, Viguié C, Wimel L, Couturier-Tarrade A, Chavatte-Palmer P. Maternal parity affects placental development, growth and metabolism of foals until 1 year and a half. Theriogenology 2018; 108:321-330.



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