Short Communications

Causes of equine abortion, stillbirth and neonatal death in central Italy

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ABORTION, stillbirths and neonatal deaths are an important source of economic loss for an equine industry. Many of the infectious disease agents that have been implicated are important from the viewpoint of international trade (Butler and others 2011). Having knowledge of the likely presence of specific infectious agents in an area is very useful for clinicians when attempting to identify the cause of an abortion and when assessing the predictive value of a diagnostic test.

As reported in previous studies (Hong and others 1993, Butler and others 2011, Laugier and others 2011), the causes of abortion can change over time, as a reflection of improved diagnostic capability or because of background differences in at-risk equine populations. Moreover, regional differences can also influence the cause and frequency of pregnancy loss (Giles and others 1993, Hong and others 1993, Tengelsen and others 1997, Smith and others 2003, Butler and others 2011, Laugier and others 2011). Monitoring and surveillance of the causes of fetal loss over time is very important.

The aim of this study was to investigate the principal causes of abortion, stillbirth and neonatal mortality in horses in central Italy, with particular emphasis on those of infectious aetiology.

Abortion was defined as fetal loss before 300 days' gestation, stillbirth as delivery of a dead foal after 300 days of gestation and neonatal mortality as foals that die within seven days of birth. A total of 67 abortions, 22 stillbirths and 14 cases of neonatal mortality were examined in this study. These represented voluntary submissions by veterinarians to the Diagnostic Laboratory, Faculty of Veterinary Medicine, University of Perugia, between November 2004 and July 2011. Only three cases were investigated in 2004. Cases came from over 31 farms and included thoroughbreds, standardbreds, warmbloods, Maremmano horses, Italian TPR agricultural horses, Arabians and Shetland ponies. Most cases were received between January and June (n=75, 73 per cent). The gestational age of aborted fetuses was three to five months (10 cases; 15 per cent), six to eight months (34 cases; 51 per cent) and nine to 10 months (23 cases; 34 per cent). The placenta was available in 81 cases (79 per cent); it was the only specimen submitted from seven abortions. The vaccinal status for equine herpes virus type 1 (EHV-1) was provided for 49 of the aborting mares. Over the three years of the study, five mares experienced repeat fetal losses.

Fetuses were subjected to detailed gross examination and selected tissues were submitted for histopathological examination. Virological, bacteriological and fungal examinations were performed on samples of lung, liver, spleen, thymus, placenta and stomach contents (the latter only with respect to bacteriological examination).

All specimens were inoculated on to blood, MacConkey's and Sabouraud agars and incubated aerobically at 37°C to determine any bacteria or fungi that might be present. A bacterial or fungal infection was diagnosed if a pure or predominant growth of a specific microorganism was obtained from one or more organs and if histological lesions were present consistent with that infection.

A PCR assay for EHV-1 and EHV-4 was routinely carried out on every case (Varrasso and others 2001, Passamonti and others 2006). Starting in 2006, viral isolation was also performed on all tissue homogenates using monolayers of the RK-13 rabbit kidney (ATCC CCL 37) cell line (OIE 2011).

PCR screening for *Leptospira interrogans* sensu lato was carried out when gross or histological lesions were suggestive of this infection (Marenzoni and others 2010). PCRs for equine infectious anaemia virus (EIAV) and equine arteritis virus (EVA) were performed only when there was clinical evidence of infection in the aborting mare(s), using previous published protocols (Gilbert and others 1997, Cappelli and others 2011).

The study findings are summarised in Table 1. Cases of EHV-1 infection were recorded every year except 2004 and 2007. Positive cases came from five different farms, two of which experienced cases of EHV-1 abortion in consecutive years even though pregnant mares were regularly vaccinated against EHV-1. A total of 18 cases of EHV-1 abortion occurred in vaccinated mares.

The two abortions due to EIAV were recorded in 2006, during an epidemic in Italy.

Salmonella species isolated from two cases of abortion were typed as *Salmonella enterica* subspecies *enterica* serovar Bareilly and *S enterica* subspecies *enterica* serovar Abortusequi.

The cause of abortion was identified in 66 of 103 cases (64 per cent). Of these, infectious agents including co-infections, accounted for 50 of 66 (76 per cent) of the diagnoses and 50 of 103 (49 per cent) of the total number of cases examined.

EHV-1 was the most frequently diagnosed cause of abortion. Other reports have also found EHV-1 as a frequent source of pregnancy loss in horses (Giles and others 1993, Hong and others 1993, Tengelsen and others 1997, Smith and others 2003, Butler and others 2011, Laugier and others 2011), in spite of vaccination against the virus or geographical location of the mares.

No cases of EVA abortion were found. This may be an underestimate of the presence of the virus as no systematic screening for this virus was carried out and diagnostic testing for this infection was restricted to mares having previous clinical evidence of EVA.

This is the first investigative study of the causes of equine abortion, stillbirth and neonatal mortality in Italy. The percentage of diagnosed cases of abortion, stillbirths and neonatal deaths in foals was lower than that of other studies (Giles and others 1993, Hong and others 1993, Tengelsen and others 1997, Smith and others 2003, Butler and others 2011, Laugier and others 2011). Non-infectious causes, in particular those frequently reported as a cause of fetal loss, need to be more fully investigated, and a more rigorous examination of placentas should be carried out to identify possible placental disorders.

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TABLE 1: Diagnostic categories of 103 cases of equine abortion, stillbirth and neonatal deaths

Diagnostic category			Cases of abortion	Cases of stillbirth	Cases of neonatal mortality	Total number (%)
Infectious causes	EHV-1		9	5	8*	22* (21.3)
	EIA		2	-	-	2 (1.9)
	Bacterial infections (septicaemia)	*Actinobacillus equuli*	4†	-	-	4 (3.8)
		Leptospira interrogans sensu lato	1‡	1‡	-	2 (1.9)
		Listeria monocytogenes	1	-	-	1 (0.9)
		Escherichia coli	-	-	1	1 (0.9)
		Klebsiella pneumoniae	5	-	2	7* (6.8)
		Salmonella enterica	2	-	-	2 (1.9)
		Agent not identified	2	1	-	3 (2.9)
	Placentitis	*Streptococcus zooepidemicus* biovar *equi*	2	-	-	2 (1.9)
		Klebsiella pneumoniae	1	-	-	1 (0.9)
		Staphylococcus aureus	1	-	-	1 (0.9)
		Enterobacter aerogenes	-	1	-	1 (0.9)
		Aspergillus ochraceus	-	1	-	1 (0.9)
		Agent not identified	1	2	-	3 (2.9)
Non-infectious causes	Umbilical cord torsion		5	1	-	6 (5.8)
	Congenital anomalies		1	-	-	1 (0.9)
	Placental dystrophic calcification		1	1	-	2 (1.9)
	Placental insufficiency		1	-	-	1 (0.9)
	Trauma (diaphragmatic rupture)		-	1	-	1 (0.9)
	Pregnancy in the uterine body		-	2	-	2 (1.9)
	Premature placental separation		-	3§	-	3 (2.9)
	Dystocia + persistent urachus		-	-	1	1 (0.9)
Non-infectious maternal disease			2	-	-	2 (1.9)
No diagnosis			28	6	3	37 (35.9)

* Two cases were complicated by co-infection with *K pneumoniae* in the lungs (included in the table) (Passamonti and others 2006) and 1 with *A equuli* (included in the table). For these cases of co-infection, the percentage exceeds 100 per cent
† Passamonti and others (2005)
‡ Marenzoni and others (2010)
§ One case was associated with EHV-1 infection, another with *Leptospira interrogans* sensu lato infection and the last one with a purulent placentitis without an identified agent. For these co-infections, the percentages exceed 100 per cent. These infections were considered mainly responsible for the stillbirths
EHV-1 Equine herpesvirus type 1, EIA Equine infectious anaemia

Because the cases included in this study were submitted on a voluntary basis, the findings may not necessarily be representative of the true incidence of the problem in the field. It is important that practitioners show greater interest in investigating the possible infectious aetiology of any cases of abortion they encounter, even those where macroscopic lesions are evident (ie, congenital defects or umbilical torsion).

A more extensive surveillance programme for equine abortion, stillbirth and neonatal mortality would be useful from the viewpoint of being able to investigate a greater number of cases, improve diagnostic techniques and monitor trends of specific infections. Many abortigenic agents are significant in terms of international trade and some from the viewpoint of public health. Emergent diseases as exemplified by the mare reproductive loss syndrome (Cohen and others 2003) are unlikely to be recognised if a programme of continued surveillance is not in place. Greater international agreement on the causes of pregnancy loss would facilitate comparison of data from different countries.

Correction notice

This article has been corrected since it was published Online First. The author's surname P. C. Proietti was incorrect. This has now been corrected to Casagrande Proietti.

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