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Diagnosis and control of Equine Infectious Anemia in a horse farm located in Buenos Aires province, Argentina

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Although Equine Infectious Anemia (EIA) is endemic in the northern area of Argentina, its epidemiological situation varies along the extent of the country, with a low prevalence in the central region and free-status in Patagonia (southern region). Argentinian legislation imposes the immediate euthanasia of horses detected as positive by agar gel immunodiffusion test (AGID). This report describes the consequences of EIA virus infection in a farm dedicated to the production of crossbreeding horses (Criollo, Arabian and Quarter horse) located in Baradero, 150 km northwest of Buenos Aires. This horse population (n: 124) had not been tested for EIA in the last 3 years, and there was no EIA control on animals admitted into the farm. The observation that motivates the EIA screening was the death, with no attributable causes, of 10 horses during last year. On March 21st 2015, the adult population of horses was tested for EIA by AGID. Twenty-four out of 109 (22%) horses were found positive. The Animal Health Authorities (SENASA) were notified and these animals were immediately euthanized, and the farm quarantined. After this, three additional AGID tests were conducted on adult horses, and other three on the whole population (including foals at foot). The obtained results are shown in the following table.

	March 21 st	May 15 th	June 23 rd	July 30 th	August 28 th	September 17 th	October 5 th
Horses tested	Adults				Adults and foals at foot		
	109	83	67	68	83	82	82
AGID Positives	24	20	5	3	1	0	0
Prevalence	22%	24%	7%	4%	1%	0%	0%

As result of EIA infection, 53 horses in all (43% of total population) had to be euthanized. The 29 horses which tested positive during the six-month period of additional testing either contracted the infection during this period or had been already infected when introduced into the farm. The inappropriate handling, with horses being either ignored in one of the tests or introduced into the farm during quarantine (shown by the incongruity in the number of horses tested in each occasion), could be the reason for the long time taken to control the infection. Even though all horses resulted negative in the last two tests, the farm still remains quarantined; another control 60 days after, with negative results, is required. The present work illustrates the devastating consequences of EIA infection, as it has brought along important economic losses for the owner, due both to the loss of positive EIA horses, which have been euthanized, as to the restrictions for the movement and sales of horses from this breeding farm. Moreover, this report emphasizes the importance of promoting EIA surveillance in the area considered as "low prevalence", considering this area is where the most valuable horses are bred in Argentina. It is also necessary to raise awareness that it is mandatory to strictly comply with legislation when dealing with EIA infection, which implies control of all horses (even foals on foot) in the facility at the same time and prohibition of entry/exit of new individuals if untested.

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Identification of Equine arteritis virus immunodominant B cell epitopes using a peptide microarray

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Equine arteritis virus (EAV) is the causative agent of equine viral arteritis (EVA), a respiratory and reproductive disease of equids. Affected adult horses make a complete recovery, but a percentage of infected stallions may become long-term carriers. The genome of EAV has a length of 12.7kb and contains seven open reading frames (ORFs). ORFs 1a and 1b represent seventy-five percent of the viral genome and code for replicase and polymerase activities. ORFs 2-7 are nested and code for the structural proteins of the virus. EAV is widely distributed in equine populations around the world and although there is only one serotype for EAV, considerable variation exists between field strains. At present, the serum neutralisation test (SNT) is the principal serological assay used to detect EAV, but the SNT is expensive and time consuming to perform. Additionally, several laboratories have reported problems relating to serum cytotoxicity in lower serum dilutions. To overcome these limitations, considerable efforts have been undertaken to develop an ELISA, but with limited success so far. The antibody response of an individual horse differs depends on its host MHC (ELA) genotype, the EAV strain, and the duration/type of infection. A peptide microarray was developed using the commercially available PEPperCHIP® microarray platform to identify immunodominant B cell (antibody) epitopes of EAV. The whole EAV strain Bucyrus amino acid sequence was transformed into a total of 625 peptide overlapping peptides that were syn-

thesized and spotted in duplicate onto a microarray slide. A panel of 28 field samples representing a selection of sera generated against a variety of known EAV genotypes was tested using the microarray. Of the 625 peptides, 97 peptides (15.52%) showed reactivity with the EAV positive samples. No single peptide was detected by all the positive serum samples. Seven peptides repeatedly showed reactivity above the cut-off and were considered to have diagnostic potential. Five of these peptides were within the immunodominant GP5 protein and two within the replicase polyprotein region NSP2 and NSP10 located in ORF1. Based on the results obtained in this study, a peptide ELISA offers a promising approach as an alternative to the SNT for the detection of antibodies to EAV. Further development and optimization is required to determine if the peptide candidates identified through this study offer the sensitivity and specificity required for the diagnosis of EAV.

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Performance of the iELISA in horses with long term guttural pouch carriage of *Strep equi equi*

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