VETERINARY VIROLOGY - VV

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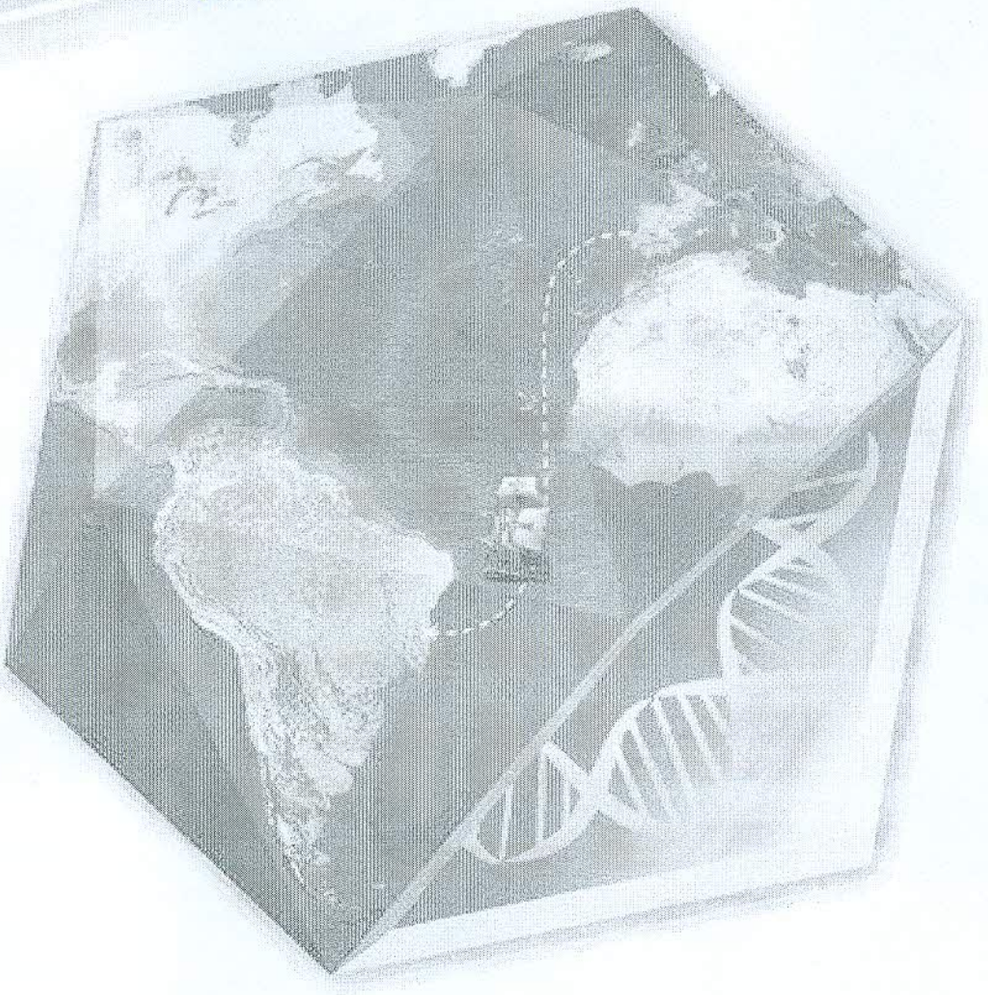
(Repro - 3 resumos)



genotypes is essential to a better understanding of rotavirus epidemiology. Financial support: FAPESP 2011/00870-0

VV132 - ORF VIRUS INFECTION IN GOAT HERDS OF SEMIARID REGION, BAHIA, BRAZIL.

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Contagious pustular dermatitis is a viral disease of worldwide distribution, caused by ORF virus belonging to the family Poxviridae. The virus causes acute pustular lesions in sheep and goats on the muzzle and lips although lesions within the mouth, affecting gums and tongue can occur. Human infection can occur among workers exposed occupationally (zoonosis). In Bahia the frequency of ORF virus infection seems to be higher than that officially reported. This study is the first report on the ORF virus identification and laboratory confirmation in herds of goats in Bahia. It was reported the occurrence of an epithelial disease in goats in the locality of Araci. The lesions were characterized by the occurrence of scabs around the labial fold or being restricted to the udders. These scabs and crusts were collected and sent to the Laboratory of Virology of the Institute of Health Sciences, Federal University of Bahia. The material from the lips and teats were submitted to tripsine 0.025% treatment and subjected to DNA extraction or the scabs were macerated with sterile sand in PBS to isolament in cell culture. The DNA obtained was submitted to the technique of polymerase chain reaction (PCR) to amplify the genes ORFV 011 (B2L, 1022 base pairs [bp]) and ORFV 059 (F1L, 1062bp). All of the samples showed positive PCR reaction and the isolament in ovine kidney cell cultures confirmed the ORF virus infection of in the goat herds of Bahia state. Financial Support: FUNDECI-Banco do Nordeste.

VV153 - GENETIC CHARACTERIZATION OF HEPADNAVIRUS IN PIGS DESTINED FOR HUMAN CONSUMPTION IN RIO DE JANEIRO, BRAZIL.

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The hepatitis B virus (HBV) belongs to the Hepadnaviridae family, which can be found in both mammals and birds. This family includes HBV that infects humans and non-human primates, including chimpanzees, gibbons, gorillas, orangutans and woolly monkeys, and HBV-related viruses circulating in rodents, such as woodchuck and squirrels, and avian hosts, like ducks, geese, herons, storks. Increasing the list of putative hosts, recent studies confirm the presence of serological markers of HBV in swine herds in China and the molecular detection of hepadnavirus in serum samples from swine herds in Santa Catarina State, Brazil. The aim of this study was to genetically characterize the hepadnavirus in bile samples from domestic pig herds in Rio de Janeiro, Brazil. For this purpose, 36 domestic pigs from slaughterhouses located in Petrópolis, Itaocara and Itaperuna (North and Hill region of Rio de Janeiro State, Brazil) were screened for Hepadnavirus-DNA. Analyses were performed in bile samples after DNA viral extraction (QIAamp DNA Mini Kit, Qiagen®), partial genome amplification and genome sequencing. From 36 bile samples analyzed, 4 samples showed a positive result for the semi-nested PCR specific for HBV ORF S (972 bp). Futhermore, phylogenetic reconstruction using the same partial nucleotide sequences of ORF S showed a close relationship of hepadnavirus strains from pigs and hepadnavirus nucleotide sequences from non-human primates (from 77.3 to 85.2%), rodents (from 55.6 to 59.2%) and birds (from 39.6 to 42.4 %) Besides, these data also reveal that distinct strains might be responsible for the infection in domestic pigs. Thus, additional studies are needed to determine a probable infectivity of this agent, as well as evaluating possible epidemiological risks. Financial support: IOC/Capes Area: Animal Virology

VV164 - MULTIPLEX PCR FOR SIMULTANEOUS DETECTION OF FIVE SINGLE-STRANDED SWINE DNA VIRUSES

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Porcine parvovirus (PPV1) and porcine circovirus type 2 (PCV2) cause clinical disease in swine. Porcine parvovirus type 4 (PPV4), torque teno virus 1 (TTV1) and torque teno virus 2 (TTV2) circulate among swine in association with PPV1 and PCV2 infections, although their epidemiology and pathology in pigs remains unclear. In order to detect these five DNA viruses from pigs in the same reaction, a multiplex polymerase chain reaction (PCR) was designed. Each DNA target produced a specific amplicon of 561bp (PPV1), 440bp (PPV4), 341bp (TTV2), 284bp (PCV2) or 101bp (TTV1). Amplicons were cloned into pCR4 plasmid (TOPO TA Cloning Kit, Invitrogen) and purified using the PureLink Quick Plasmid Miniprep Kit (Invitrogen). All cloned amplicons were verified by sequencing, quantified using a ND-1000 spectrophotometer and 10 serial dilutions of 1.2×1010 up to 1.2×100 DNA copies/μL were done. The multiplex PCR was performed in a 30μL reaction and carried out with all five primer pairs with optimized parameters. The reaction contained 4mM MgCl2, 1.5× PCR Buffer, 0.4mM dNTP, 0.05μM TTV1, TTV2 and PCV2 primers, 0.16μM PPV1 primer, 0.4μM PPV4 primer and 1.8U Platinum Taq DNA Polymerase (Invitrogen). The PCR cycling conditions consisted of an initial denaturing at 95°C for 30s followed by 35 cycles at 95°C for 1 min, 60°C for 1 min and 72°C for 1 min followed by a final extension at 72°C for 10 min. The amplicons were analyzed by electrophoresis through a 2% agarose-TBE gel and stained with ethidium bromide. The sensitivity of the multiplex PCR was 1.2×103 DNA copies/μL. The specificity of the multiplex PCR was tested using influenza A virus and PCV1 and no amplification was observed. Therefore, the multiplex PCR developed in this work showed to be a rapid, sensitive, specific and cost-effective method for the detection of five swine viruses and can provide a useful tool for clinical analysis and epidemiology of these viruses in swine herds. Financial support: Embrapa (Process nº. 02.11.10600-03)

VV178 - EXPERIMENTAL EVALUATION OF VACCINE-INDUCED SEROCONVERSION OF AVIAN LIVE VACCINES SUBMITTED TO THE OFFICIAL QUALITY CONTROL DURING SEPTEMBER 2012 TO MAY 2013

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The prophylaxis of many avian diseases is mainly based on active immunization through the employment of live vaccines. One of the most important test on the quality control of vaccines is the seroconversion. Seroconversion is carried out to verify if detectable specific antibodies to the antigen were produced, after immunization. The Avian Health Unit from Lanagro-SP performs the Quality Control of avian live vaccines commercially available in Brazil. The objective of this study was to evaluate experimental seroconversion assays carried out from Sept.2012 to May 2013. Seroconversion test was performed using two glasses of live vaccines. Vaccines were reconstituted in phosphate buffer saline, inoculated in SPF chicks according manufacturer's recommendations and kept in BSL3-isolators. Around 21-28 days after immunization chicks were bled via cardiac puncture, serum was obtained and serological tests (HI and/or ELISA) were performed. Fifty-six batches of live vaccines were tested: 16 batches of infectious bursal disease (IBD), 14 batches of infectious bronchitis (IB) disease, 6 batches of Newcastle disease (ND), 5 of combined vaccines (4 batches with ND+ IB fractions and 01 Marek + IBD), and 5 of complexes vaccines (IBD + antibodies). The results showed that ND vaccines had geometric mean titers (GMT)/HI = 22 to 84.4 and GMT/ Elisa = 3,581. IBD vaccines had GMT = 3,274 to 14,594; IBD vaccines produced with strong strains had biggest values of GMT. The results of complexes vaccines showed GMT = 4,587 to 5,917. Considering results obtained with ND, IBD and complexes vaccines, all batches have been approved. However, 85.7% of IB vaccines (GMT = 0 to 470.6) and 80% of the combined vaccines that have the fraction IB, were failed. These results show the importance of seroconversion test for the quality control of avian live vaccines used in the country to assure that good veterinary vaccines are available for commercial use. Financial Support : Ministério da Agricultura, Pecuária e Abastecimento

VV182 - IMPLEMENTATION OF PCR AND RT-PCR FOR DIAGNOSIS OF FELINE HERPESVIRUS (FEHV) AND FELINE CALICIVIRUS (FCV) INFECTION IN RIO DE JANEIRO.

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Feline calicivirus (FCV) and herpesvirus (FeHV) are the main agents of conjunctivitis and upper respiratory tract disease in domestic young cats and these agents are widespread in the domestic cat population, despite the use of the vaccines. Previous reports in southern part of Brazil have shown that FCV and FeHV-1 were detected in about 39% and 53% of the cats, respectively. However, the occurence of FCV or FeHV have not been described in the State of Rio de Janeiro yet. In this study, conjunctival swabs were collected from 16 unvaccinated kittens (up to one year) with clinical signs of conjunctivitis from 3