

analysed to detect the presence of astrovirus using RT-PCR. In total, 38 bats samples belonging to thirteen species were studied. The samples were submitted to RNA extraction followed by RT-PCR. As results, three positive samples (11%) were found in only one species (*Tadarida brasiliensis*). Those positive samples were collected in urban areas, while the presence of astrovirus was not identified in samples from natural environment. Possibly, the anthropic environment may contribute to the presence of the virus, since previous studies showed that bat astroviruses may be related to human astrovirus, but these possibility will be further studied. FINANCIAL SUPPORT: CAPES, CNPq, FAPERGS and Propesq/UFRGS.

VV207 - DETECTION OF AVIAN GROUP F ROTAVIRUS IN FECAL SAMPLES OF BROILER CHICKENS IN PARA STATE, BRAZIL

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Rotavirus (RV) is a major viral agent of enteric disease in humans and animals. They are non-enveloped viruses with genome divided into eleven segments of double-stranded RNA that encodes 12 proteins. The VP6 protein is conserved and an excellent target for laboratory diagnosis as well as being determinant of species/groups of RV these are classified into eight (A-H). The group F rotaviruses (RVF) infect only birds causing damage to health and consequently losses to the poultry market. Presently, there are few data in the literature on the RVF particularly using molecular methodologies such as the reverse transcriptase-polymerase chain reaction (RT-PCR). In this context, this study aimed to detect RVF in broilers in Para state, Brazil, during 2008 to 2011. A total of 85 pools of fecal specimens of broilers were collected in 37 farms located in eight different municipalities in the metropolitan region of Belém (Belém, Ananindeua, Benevides, Castanhal, Santa Isabel do Pará, Inhagapi, Santa Bárbara do Pará e Santo Antônio do Tauá). Viral genome was extracted from stool suspensions and subjected to RT-PCR using a specific primer pair designed for the gene encoding VP6 protein of RVF (RF6F / RF6R). The positive samples were sequenced using the same primers in RT-PCR. RVF positivity was detected in 9.4%

(8/85) of pools. Samples from five of eight municipalities were positive for RVF with infections spread in 7 of 37 (18.9%) farms. The higher rate of infection was related to the age of 16-30 days. This study is one of the pioneers to detect RVF by RT-PCR, as well as adding information about the occurrence of RVF in Brazil, providing more representative data about this group of RV. FINANCIAL SUPPORT: FAPESPA; CNPQ.

VV229 - LONG-TERM CIRCULATION OF INFLUENZA A VIRUSES IN SWINE IN BRAZIL

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Influenza A viruses (IAVs) circulating in swine present important economic concerns for the swine industry and a pandemic threat for humans. Although Brazil hosts one of the largest swine populations in the world, there has been little evidence prior to 2009 of IAV circulation in Brazilian swine herds. Following the detection of pandemic H1N1 (H1N1pdm) IAVs in pigs in Brazil in 2009, surveillance efforts increased. Screening of 1440 nasal swab samples was carried out by RT-qPCR assay. Thirty-nine positive samples were submitted to virus isolation. Genetic sequencing was performed by ABI 3130xl and Illumina MiSeq. Five H1N2, four H3N2 and seven H1N1pdm IAVs, collected from swine in different Brazilian states during 2009-2012, were sequenced and analyzed. Nucleotide alignments were generated for five data sets: 'H1s' (human seasonal virus-like), 'H1p' (pandemic virus-like), H3, N1p (pandemic virus-like) and N2, including other related human and swine viruses, collected globally, as background. The four H3N2 viruses from Brazilian swine are monophyletic (100% bootstrap) and closely related to human seasonal viruses from the late 1990s. The five H1N2 viruses are also monophyletic (64% bootstrap) on the H1 tree, and are closely related to seasonal H1N2 viruses that circulated in humans during 2001-2003. The lower support for this clade appears to be driven by the early divergence of the Brazilian H1 viruses into two distinct sub-clades. The Brazilian swine IAVs are not monophyletic on the N2

phylogeny. Five Brazilian swine viruses of the H1N2 and H3N2 subtypes belong to one N2 clade (97% bootstrap) and two H1N2 belong to a second N2 clade (100% bootstrap). Both clades are closely related to human seasonal H3N2 viruses from the late 1990s. Therefore, Brazilian swine viruses of the H1N2 subtype that contain H1 segments related to human seasonal H1N2 viruses have acquired a different N2 of human H3N2 origin via two different reassortment events. The seven viruses of human pandemic H1N1 origin also were not monophyletic on the H1 or N1 tree, indicating that these viruses are the result of multiple separate human-to-swine introductions of the H1N1pdm virus, rather than clonal expansion of a single introduced lineage. The co-circulation of multiple antigenically diverse influenza virus lineages of the H1N1, H1N2, and H3N2 subtypes introduces new challenges for the control of influenza in Brazil's swine herds, including design of cross-protective vaccines. FINANCIAL SUPPORT: EMBRAPA (PROCESS Nº. 02.11.10600-01)

VV260 - EVIDENCE OF ORTHOPOXVIRUS CIRCULATION AMONG URBAN DOMESTIC CATS, MINAS GERAIS, BRAZIL

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Cessation of smallpox vaccination occurred 36 years ago in reason of its eradication. Because of this, several outbreaks involving other orthopoxviruses have been reported worldwide, such as Monkeypox virus which is endemic in Africa and Cowpox virus associated to human and animal cases in Europe, specially having cats as hosts. In Brazil, Vaccinia virus is a causative agent of an exanthematous disease, always affecting dairy cattle and humans and related to rural environment. Furthermore, other mammalian species are known to be infected, such as equids, monkeys and wild rodents. Taking into account that cats are well known to transmit Cowpox virus in European urban areas, our goal was to investigate Orthopoxvirus circulation among cats in a Brazilian urban environment and its implication for epidemiological studies. Whole blood,

plasma and serum samples were analyzed from 78 domestic cats that live in an urban area from state of Minas Gerais. Plaque reduction neutralizing test was applied in serum samples to check anti-Orthopoxvirus neutralizing antibodies. Viral DNA was accessed using phenol, chloroform and isoamyl alcohol method and real time PCR targeting vgf and ha genes were also applied. Most animals are female (55.1%) and age range from 5 months to 11 years. It was found 13 seropositive samples (16.7%), with antibodies titers ranging from 100 to 1600 neutralizing units per ml. Molecular analysis of seropositive cats showed 7 positives for vgf gene and 4 for ha. Poxviruses are ubiquitous among mammals and the host spectrum is wide. Although in Brazil occurrence of poxviruses are, until now, restricted to rural environment involving dairy cattle and humans, investigations conducted in urban areas highlights the importance to clarify epidemiological chain of this emerging infectious disease. Since smallpox eradication, vaccination is discontinued worldwide increasing the number of immunologically unprotected people. These data also reinforce the impact of viral spread to urban environments affecting vulnerable populations. FINANCIAL SUPPORT: CAPES, CNPQ, FAPEMIG, PRPQ UFMG, PPG-MICROBIOLOGIA UFMG.

VV303 - DEVELOPMENT AND STANDARDIZATION OF A MULTIPLEX RT-PCR FOR THE DETECTION THE PRINCIPAL RESPIRATORY VIRUS IN BIRDS

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The avian Metapneumovirus (aMPV), the Newcastle disease virus (NDV), Influenza A virus (AIV) and infectious bronchitis virus (IBV) are the most important respiratory virus pathogens that affect breeding hens and broilers, with high importance to the poultry farms, both in terms of economic losses, impact in the exportation as the possibility of transmission between birds species. The aim of the present paper proposes the development, evaluation and standardization of a Multiplex RT-PCR, in order to assess the purity of avian vaccines. Reference virus: aMPV subtypes A and B; NDV-La Sota; AIV subtype H5N2; IBV-M41 were used for standardization of the assay and commercial vaccines