

DETECTION OF CANINE DISTEMPER VIRUS BY RT-PCR IN SERUM, LEUKOCYTES, CEREBRO-SPINAL FLUID (CSF), AND URINE IN DOGS WITH DISTEMPER ENCEPHALITIS: 11 CASES. T.B. Saito, A.A. Alfieri, A.F. Alfieri, S.N.E. Beloni, H.S.A. de Moraes. Universidade Estadual de Londrina, Londrina, Paraná, Brazil

We prospectively compared serum, leukocytes, CSF, and urine for diagnosing distemper encephalitis by RT-PCR in dogs. Poorly vaccinated dogs with progressive multifocal central nervous system disease from our Hospital population were selected for the study. Twenty dogs fulfilled the criteria for inclusion. Eleven of them, including 7 with myoclonus, were diagnosed as having distemper. Distemper encephalitis was confirmed based on a positive RT-PCR in the CSF. When CSF was not available, distemper was diagnosed whenever RT-PCR was positive in serum, leukocytes, and urine. One dog was negative in the CSF, but positive in all other samples. This dog was considered to have distemper. One dog was positive only in the urine and considered negative. The remaining negative dogs were negative in all materials tested. The RT-PCR amplified a fragment from the N gene of the nucleic acid of canine distemper virus (CDV). RNA was extracted using guanidinium thiocyanate. The thermocycle profile consisted of 40 cycles 94 C for 1 minute, 59.5 C for two minutes, and 72 C for 1 minute. The resulting fragment had 287 base pairs. We have previously shown that this RT-PCR is specific for the CDV, but also amplifies the measles virus. CDV and measles virus were differentiated through fragment analysis using a HINF1 enzyme.

RT-PCR for CDV was run in samples of serum, urine, and leukocytes from all dogs. Leukocytes were separated using a percoll gradient of 1.08. The majority of cells obtained were lymphocytes with small amounts of monocytes and neutrophils. RT-PCR was also run in CSF samples from 6 distemper dogs, being positive in 5. The dog considered positive despite the negative RT-PCR in CSF had viral fragment amplified in serum, leukocytes, and urine. Four dogs were positive only in CSF and urine. All distemper dogs had positive results in the urine, but only 7 were positive in leukocytes and serum. In conclusion, serum and leukocytes do not appear to be very sensitive for diagnosing naturally-occurring distemper encephalitis in dogs. Urine was the only fluid found positive in all dogs, suggesting its usefulness as a routine screen for dogs in which infection by CDV is a potential differential for the CNS signs. These results also suggest that elimination of the virus in the urine may play an important role in cross-contamination and maintenance of the virus in the environment