DEVELOPMENT OF SEMI-NESTED-RT-PCR TECHNIQUE FOR THE DETECTION OF NEWCASTLE DISEASE VIRUS IN PIGEONS CAPTURED IN JABOTICABAL-SP, BRAZIL

(DESENVOLVIMENTO DA TÉCNICA DE SEMI-NESTED-RT-PCR PARA A DETECÇÃO DO VIRUS DA DOENÇA DE NEWCASTLE EM POMBOS CAPTURADOS EM JABOTICABAL-SP, BRASIL)

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The Newcastle disease virus (NDV) is the causative agent of an important disease of domestic and wild birds and poses a threat to the poultry industry. Pigeons (Columba livia) are susceptible to NDV and considered potential spreaders of the virus. Therefore, pigeons have an important role in the epidemiology of this disease that can impact negatively the poultry industry. This study aimed to investigate the presence of NDV on cloacal swabs of 101 pigeons captured in Jaboticabal. The molecular technique is based on semi-nested RT-PCR and targets the amplification of the coding region of the Fusion protein 196 bp. We assessed four combinations of external and internal primers in the semi-nested RT-PCR technique by testing serial dilutions of the samples in chorion-allantoic fluid (CAF) infected with LaSota strain of NDV. The combination of external and internal primers that yielded higher analytical sensitivity was used in the analysis of cloacal swabs of pigeons. The results showed that the best combination of external and internal primers presented a detection threshold in semi-nested RT-PCR up to 10⁻¹⁰ dilution of the CAF with NDV suspension. The analytical specificity of the primers was also evaluated using CAF suspensions infected with four viruses heterologous to the NDV, of which no amplified product was generated. The results showed that of 101 samples of pigeon cloacal swabs analyzed, 9 (8.9%) were positive for NDV. In conclusion, the semi-nested RT-PCR technique developed in this study was efficient for specific detection with high sensitivity to NDV in both samples, CAF and pigeon cloacal swabs, indicating its great potential for application in direct diagnosis of NDV in different types of biological samples from poultry.

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