COMPARISON OF VIRUS ISOLATION AND POLYMERASE CHAIN REACTION FOR DIAGNOSIS OF PESTE DES PETITS RUMINANTS

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Summary. — Oculonasal swabs and tissue samples collected from peste des petits ruminants (PPR) suspected sheep and goats were tested for presence of the virus of peste des petits ruminants (PPRV) or its RNA by reverse transcription–PCR (RT-PCR) and virus isolation (VI). Of 44 samples 31.8% and 40.9% were positive by VI and RT-PCR, respectively. The RT-PCR-positive samples were subjected to the nested PCR. Three of six samples positive by RT-PCR but negative by VI were negative by the nested PCR. The specificity and accuracy of the nested PCR were higher than those of the RT-PCR although the sensitivity of both tests were similar. Nucleotide sequencing of one nested PCR product revealed a 92% homology with the sequence available in the GenBank (Acc. No. Z37017).

Key words: peste des petits ruminants; reverse transcription–polymerase chain reaction, virus isolation, nested PCR

Introduction

PPR is an acute, highly contagious and fatal disease of small ruminants. The causative agent, PPRV (genus Morbillivirus, subfamily Paramyxovirinae, family Paramyxoviridae), is closely related to its counterpart — the rinderpest virus (RPV) (Saliki et al., 1993). Generally, RPV causes the disease in large ruminants, such as cattle and buffalo, while PPRV in small ruminants such as goats and sheep. However, in India, RPV causes clinical and subclinical infection in small ruminants too (Taylor, 1986), which can then be transmitted to cattle causing a more severe disease (Anderson et al., 1990) while PPRV is known to cause only a subclinical infection in cattle (Diallo et al., 1989).

Shaila et al. (1989) first reported PPRV in India and several PPR outbreaks have been reported since then in almost all the states of the country (Chandran Choudhary et al., 1995; Kulkarni et al., 1996; Nanda et al., 1996). As differential diagnosis of rinderpest and PPR by host species and clinical signs is not always correct, laboratory diagnosis is necessary, especially when small ruminants may be a source of infection of cattle. The laboratory techniques available to differentiate these diseases include virus neutralization (Taylor, 1979; Chandran et al., 1995), hybridization with cDNA probes (Diallo et al., 1989; Pandey et al., 1992), immunocapture enzyme-linked immunosorbent assay (ELISA) (Libeau et al., 1994), immunofluorescence (IF) test (Sumption et al., 1998) and RT-PCR (Forsyth and Barrett, 1995). RT-PCR can also produce a DNA product which can be sequenced.

Before using RT PCR in routine diagnostic testing of field samples, it has to be compared with the “gold standard”, the VI. The latter cannot always be done as a routine diagnostic assay owing to its time-consuming, cumbersome procedure and requirement of cell culture facilities. Forsyth and Barrett (1993) have successfully used RT-PCR to diagnose PPRV and differentiate it from RPV in samples.