ASSESSMENT OF RECENT DIAGNOSTIC TECHNIQUES FOR DIAGNOSIS OF BACTERIA CAUSING ABORTION IN CATTLE

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ABSTRACT

Brucellosis, Leptospirosis, Mycoplasmosis and Listeriosis are important zoonosis and represent a significant cause of reproductive losses in animals around the world, especially in Mediterranean countries and Egypt. Aiming at improvement in the diagnostic scheme, a rapid method for the simultaneous detection of these microorganisms in different clinical samples using a multiplex conventional and a SYBR green real-time PCR has been developed. The m PCR has been standardized by using 4-pairs of primers to amplify 31kDa gene encoding protein in *Brucella* spp., *lig* gene in pathogenic *Leptospira*, *hly*A gene in *Listeria monocytogenes* and *16S* rDNA in *Mycoplasma* spp. This study was applied to 166 different clinical samples (115 milk samples, 10 semen samples, 6 tissue samples, 15 blood samples, 5 vaginal swabs and discharges and15 fetal fluids and fetal organs from aborted foeti). The results of conventional PCR showed an expected band at 223bp, 468p, 370 bp and 270bp were obtained from *Brucella* spp., pathogenic *Leptospira*, *Listeria monocytogenes* and *Mycoplasma bovis*, respectively; while the real time PCR assay revealed a specific dissociation peak at Tm=79.0ºC, 80.0ºC, 85.2ºC and 88.0ºC (± 0.5) for *Listeria*, *mycoplasma*, *brucella* and *Leptospira* respectively. In conclusion the high throughput, time saving and cost effective of m- PCR method developed in this study, could provide a powerful tool for simultaneous, rapid and reliable detection of these microbial agents in different clinical samples.

Key words: Abortion, Cattle, Bacteria, m-PCR, RT-PCR

Contents

List of Tables

`List of Figures