**Minimal inhibitory concentration of azithromycin in *Rhodococcus equi* strains isolated from foals**

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*Rhodococcus equi* (*R. equi*) is a facultative intracellular Gram-positive organism that causes chronic pyogranulomatous pneumonia and enteritis in foals, mainly from 1 to 6 months-old (Giguère and Prescott, 1997). It is less frequently associated with other clinical signs in domestic species (Radostits et al., 2000; Takai et al., 2003). During the last years, the organism has emerged as an important opportunistic pathogen in immunosuppressed patients, especially those infected by the immunodeficiency virus (Severo et al., 2001). In Brazil, rhodococcosis is considered to be one of the most important diseases in foals (Ribeiro, 2005). Recently, it has been described in human, including patient with aids (Severo et al., 2001). *R. equi* expresses different virulence factors, including intracellular maintenance in macrophages, presence of virulence-associated plasmids, and resistance to conventional antimicrobial therapy, which may lead to chronic infections difficult to be treated, and high mortality rates in foals (Takai, 1997; Ribeiro et al., 2002).

Improved survival rates in the treatment of foals have been obtained in last years with the association between rifampin and erythromycin (Giguère and Prescott, 1997). However, erythromycin presents a variable rate of absorption by oral route, requiring multiple daily doses and showing high incidence of undesirable collateral effects (Stratton-Phelps et al., 2000). Recently, azithromycin has been investigated as an alternative drug to erythromycin, due to high bioavailability and chemical stability, and because it reaches higher concentrations in tissues and alveolar macrophages (Tavares, 2000; Davis et al., 2002). The purpose of present study was to investigate the minimal inhibitory concentration of azithromycin in *R. equi* strains isolated from foals in Brazil.

A total of 42 *R. equi* strains were used in this study. They were isolated from pyogranulomatous pneumonia (40 strains) (Fig. 1), bronchiolar lavage (1 strain) and mesenteric lymphadenitis (1 strain) obtained at the domestic animal infectious diseases sector at Faculdade de Medicina Veterinária e Zootecnia – UNESP - Botucatu, SP, Brazil. The strains were isolated in defibrinated sheep blood agar (5%), incubated at 37°C for 72-96 hours under aerobiosis. *R. equi* strains were classified based on macro and microscopical morphology, presence of virulence-associated plasmids, and resistance to conventional antimicrobial therapy, which may lead to chronic infections difficult to be treated, and high mortality rates in foals (Takai, 1997; Ribeiro et al., 2002).

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The 42 \textit{R. equi} isolates and the control strain were susceptible to azithromycin in the disk diffusion method (≥18mm), based on the Performance... (1999). All the isolates and the control strain presented MIC values that demonstrated their susceptibility to azithromycin. The MIC in which 90% of the isolates were inhibited (MIC$_{90}$) by azithromycin was equal to 1.5µg/ml.

In Brazil, rhodococcosis is considered to be one of the most common disease of foals, causing severe chronic pyogranulomatous pneumonia and/or enteric signs, leading to high mortality rates. A recent study have demonstrated the high virulence of \textit{R. equi} strains isolated from foals in Brazil (Ribeiro et al., 2005).

Despite of the efficacy of the rifampin-erythromycin association for the treatment of \textit{R. equi} infections in foals, different authors have reported concerns about the resistance of \textit{R. equi} to these drugs (Precott and Sweeney, 1985; Giguère and Prescott, 1997; Ribeiro et al., 2002). Moreover, several complications have been reported with the long-term use of erythromycin in foals, including anorexia, hyperthermia, tachypnea, bruxism, colic and diarrhea (Giguère and Prescott, 1997; Stratton-Phelps et al., 2000). Due to the undesirable collateral effects described in long-term use of rifampin and erythromycin, coupled with the development of resistant strains, other drugs have been studied as alternatives for the treatment of \textit{R. equi} infections in foals, especially azithromycin.

Azithromycin is an antimicrobial of the macrolide group. It has several qualities, such as the possibility of oral and intravenous administration, high stability in acid pH, high concentration in tissues and macrophages, lower incidence of gastrointestinal adverse effects, good oral bioavailability and broad spectrum against Gram-negative and mainly Gram-positive microorganisms, including \textit{R. equi} strains from human and animal origins (Davis et al., 2002).

In recent years, different \textit{in vitro} studies have evaluated the susceptibility of \textit{R. equi} strains isolated from foals using the disk diffusion method. However, few studies have investigated the minimal inhibitory concentration of the \textit{R. equi} isolates (Jacks et al., 2003).

In Brazil, the \textit{in vitro} susceptibility test of 39 \textit{R. equi} strains isolated from 37 pulmonar and two enteric infections in foals was performed, using the standard disk diffusion method. They showed 100.0% sensitivity to azithromycin (Ribeiro et al., 2001). Jacks et al. (2003) reported MIC$_{90}$ in 64 \textit{R. equi} strains isolated from foals as equal to 1.0µg/ml. Similarly, in the present study, all the 42 isolates and the control strain were susceptible to azithromycin in the disk diffusion method, and showed a MIC$_{90}$ value ≤1.5µg/ml, demonstrating the great effectiveness of azithromycin in Brazilian \textit{R. equi} isolates.
Results observed in the present study suggest that azithromycin can be an effective drug in the association with rifampin in rhodococcosis in foals and a possible alternative to erythromycin in the treatment of this infection.

Keywords: foal Rhodococcus equi, azithromycin, minimal inhibitory concentration

RESUMO

O perfil de sensibilidade microbiana e a concentração inibitória mínima (MIC) da azitromicina para 42 cepas de Rhodococcus equi isoladas de potros, no Brasil, e em uma cepa-controle, foi avaliado, respectivamente, pelos métodos de difusão com discos e E-test. A azitromicina apresentou 100% de efetividade in vitro para todas as cepas em ambos os testes. As cepas de R. equi apresentaram MIC90 para azitromicina em valores ≤1.5µg/ml. Este estudo mostra a alta efetividade da azitromicina em linhagens de R. equi isoladas no Brasil, sugerindo o uso dessa droga como alternativa na terapia da rodococose em potros.

Palavras-chave: potro, Rhodococcus equi, azitromicina, concentração inibitória mínima

REFERENCES


