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In Vitro Activities of Enoxacin, Enrofloxacin, Sparfloxacin, and Ciprofloxacin against *Escherichia coli* Strains Isolated from Diarrheic Lambs and Kids

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The in vitro activities of four fluoroquinolone compounds were tested against 92 *Escherichia coli* strains of ovine and caprine origin under aerobic and anaerobic incubation conditions. The four fluoroquinolones proved to be highly effective against the *E. coli* isolates tested. When bacteria were cultured anaerobically, at least fourfold increases in the MICs of enoxacin for the strains occurred and no detectable changes in enrofloxacin, sparfloxacin, and ciprofloxacin MICs occurred.

The fluoroquinolone class of compounds includes many extremely potent, broad-spectrum antimicrobial agents with excellent activity against many gram-negative bacteria (16). This class of antimicrobial agents has shown excellent in vitro activity against *Escherichia coli* clinical isolates of human origin (6, 15), including enterotoxigenic *E. coli* (3, 5, 12), and many have been used successfully in the prevention and treatment of traveler's diarrhea (4, 9). In veterinary medicine, fluoroquinolones have been introduced recently and few reports about their in vitro activities against bacterial clinical isolates of animal origin exist (11). In this report, we describe the results of in vitro activities of four fluoroquinolone compounds (enoxacin, enrofloxacin, sparfloxacin, and ciprofloxacin) against *E. coli* strains isolated from lambs and kids affected by neonatal diarrhea.

We also evaluated the in vitro activities of the four fluoroquinolone compounds against the same *E. coli* strains grown under anaerobic conditions. Against anaerobic species, activities of fluoroquinolones tend to be lower than against aerobic species (16). Since *E. coli* is a facultatively anaerobic bacterium, we have tested the in vitro susceptibilities of *E. coli* strains cultured under anaerobic conditions and compared them with the in vitro susceptibilities under aerobic conditions.

The study was done with 92 *E. coli* strains collected between 1989 and 1992 from diarrheic lambs and kids belonging to different flocks of the central region of Spain. Fluoroquinolone therapy has not been introduced in these flocks.

The following antimicrobial agents were provided by the manufacturers: enoxacin (Almirall), enrofloxacin and ciprofloxacin (Bayer), and sparfloxacin (Rhone). Laboratory powders were reconstituted in appropriate solvents as recommended by the manufacturers.

MICs were determined by an agar dilution method as outlined by the National Committee for Clinical Laboratory Standards (7), with Mueller-Hinton agar (Difco). Susceptibility tests under aerobic and anaerobic culture conditions were performed in the same manner, except that inocula and agar plates were cultured in room air for aerobic susceptibility tests and in GasPak jars (BBL Microbiology Systems, Cockeysville, Md.) for anaerobic tests. The plates were incubated for 24 h at 37°C, and the MIC was read as the lowest concentration of antimicrobial agent that suppressed visible bacterial growth. Reference strain *E. coli* ATCC 25922 was included as an internal control in all parts of the study.

The MICs for 50 and 90% of the isolates tested and the ranges of MICs of the four compounds for the 92 strains are shown in Table 1. The results revealed that the four fluoroquinolones tested have excellent in vitro activities against the *E. coli* isolates of ovine and caprine origin tested. When bacteria were cultured under aerobic conditions, enoxacin proved to be inhibitory for 90% of the strains at a concentration of 0.125 μg/ml. Enrofloxacin, sparfloxacin, and ciprofloxacin inhibited 90% of the strains at a concentration of ≤0.062 μg/ml. These MICs are considerably lower than MIC breakpoints indicating susceptibility at 1 μg/ml for ciprofloxacin and 2 μg/ml for enoxacin, enrofloxacin, and sparfloxacin. Thus, all the isolates were highly susceptible to the four quinolone compounds.

The in vitro activities of the fluoroquinolones tested against *E. coli* strains of ovine and caprine origin are similar to the in vitro activities against isolates of human origin reported previously (3, 15). However, Pohl et al. (11) have reported that relatively high percentages of *E. coli* isolates of bovine origin were resistant to enrofloxacin activity in vitro. A high percentage of the isolates investigated by these authors was resistant to nalidixic acid, whereas our isolates were highly susceptible to nalidixic acid. Barry and Fuchs (1) have reported that nalidixic acid-resistant strains of *E. coli* of human origin required increased concentrations of fluoroquinolones for inhibition.

When bacteria were cultured anaerobically, at least fourfold increases in the MICs of enoxacin for the strains occurred and no detectable changes in enrofloxacin, sparfloxacin, and ciprofloxacin MICs occurred (Table 1). A lowering in the pH has been reported to increase the MIC of fluoroquinolones (14). Under anaerobic culture conditions, pH may decrease and this could explain the increases in enoxacin MICs for the strains. Fluoroquinolones have been shown to have little effect against anaerobic intestinal bacteria (2, 8, 13). However, the basis for the reduced susceptibility of anaerobic organisms to fluoroquinolones is unknown. It has been suggested that anaerobic bacteria are inherently less susceptible to quinolones than

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TABLE 1. In vitro susceptibilities of 92 *E. coli* strains of ovine and caprine origin cultured under aerobic and anaerobic conditions to four fluoroquinolone compounds

Antimicrobial agent	Culture conditions	MIC (μg/ml)		
		Range	50%[a]	90%[a]
Enoxacin	Aerobic	≤0.062–0.25	≤0.062	0.125
	Anaerobic	≤0.062–1	0.25	0.5
Enrofloxacin	Aerobic and anaerobic	≤0.062–0.5	≤0.062	≤0.062
Sparfloxacin	Aerobic and anaerobic	≤0.062	≤0.062	≤0.062
Ciprofloxacin	Aerobic and anaerobic	≤0.062	≤0.062	≤0.062

[a] MIC for 50 or 90% of the strains tested.

aerobic strains because of a less susceptible DNA gyrase or that the pH of the environment in which these organisms grow affects the pK_a of quinolones, making them less active (10). According to our results, the four fluoroquinolone compounds have excellent in vitro activities against *E. coli* strains even under an oxygen-restricted atmosphere.

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