J. Dairy Sci. 99:1–13
http://dx.doi.org/10.3168/jds.2015-10807

Randomized noninferiority field trial comparing 2 first-generation cephalosporin products at dry off in quarters receiving an internal teat sealant in dairy cows

P. A. Ospina,* N. Rota,† C. Locatelli,‡ L. Colombo,† C. Pollera,† G. Giacinti,§ V. Bronzo,† A. Casula,† A. Arpinelli,# V. Brossette,‖ M. Facchi,¶ A. Patelli, A. Ruggeri,†† A. Barberio,‡‡ G. Potenza,§§ D. V. Nydam,* and P. Moroni*†[1]**

*Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853
†Dipartimento di Medicina Veterinaria, and
‡Dipartimento di Scienze Veterinarie per la Salute, la Produzione Animale e la Sicurezza Alimentare, Università degli Studi di Milano, via Celoria 10, 20133 Milan, Italy
§Istituto Zooprofilattico Sperimentale del Lazio e della Toscana Mario Aleandri, Via Appia Nuova 1411, 00178, Rome, Italy
#Practitioner, via S. Bernadette 31, 00167 Rome, Italy
‖Practitioner, via Guido Reni 96/66, 10136 Turin, Italy
¶Studio Associato Veterinari Bergamaschi via Verdi 8, 24060 Castelli Calepio, Bergamo, Italy
**Embryovet Cascina Bosco 6, 25028 Verolonuova, Brescia, Italy
††Armigio via Donatori di Sangue 36, 25020 San Paolo, Brescia, Italy
‡‡Istituto Zooprofilattico Sperimentale delle Venezie, Sez. Terr. Vicenza, 36100 Vicenza, Italia
§§Merial Italia SpA, Viale Bodio, 37, 20158 Milan, Italy

ABSTRACT

The study objective was to compare 2 commercial dry cow mastitis products at the quarter level, with concurrent internal teat sealant application, evaluating the cure risk difference, odds of a cure, odds of a new intramammary infection (NIMI) during the dry period, and risk for a clinical mastitis (CM) case between calving and 60 d in milk (DIM). A total of 590 cows (2,360 quarters) from 8 commercial dairy herds in Italy were enrolled and randomized to 1 of the 2 treatments at dry off: Cefovet A (CF; 250 mg of cephazoline; Merial Italia SpA, Milan, Italy), and Cepravin (CP; 250 mg of cephalonium dehydrate MSD Animal Health Srl, Segrate, Italy). Quarter milk samples were collected before dry cow therapy treatment at dry off, 2 to 9 DIM, and 10 to 17 DIM. Quarter milk samples from CM cases were collected during the first 60 DIM. Noninferiority analysis was used to evaluate the effect of treatment on the risk difference of a bacteriological cure during the dry period, the primary outcome. The odds of cure, developing a NIMI during the dry period, and the risk of a CM event within 60 DIM were evaluated with multivariable logistic regression and hazard analysis, respectively. The overall crude quarter-level prevalence of NIMI at dry off was 15.3%. The most common pathogen isolated from milk samples at dry-off was coagulase-negative staphylococci. Noninferiority analysis showed no effect of treatment on the risk difference for a cure between dry off and both postpartum samples, difference was 0.013. The least squares means from the multivariable model evaluating the odds of cure was 94% for CF and 95%for CP. We observed no effect of treatment on the odds for the presence of a NIMI at 2 to 9 DIM (least squares means: CF = 0.09 and CP = 0.07), nor did we note a difference in risk of experiencing a CM event between calving and 60 DIM (hazard ratio = 0.8). In conclusion, no difference was observed between the 2 products evaluated when assessing the aforementioned outcomes in quarters also receiving an internal teat sealant.

Key words: dry cow therapy, randomized noninferiority field-trial, first-generation cephalosporin, intramammary infection

INTRODUCTION

Blanket dry cow therapy (**DCT**), which refers to the intramammary infusion of all quarters of all cows at dry off with a long-acting antibiotic, is a procedure recommended by the National Mastitis Council (**NMC**) as a mastitis-control practice. The DCT is used for curing susceptible existing subclinical infections and for preventing new intramammary infections (**NIMI**), which could be acquired during the early dry period. This DCT has helped to reduce the NIMI risk from 30

Received December 22, 2015.
Accepted April 25, 2016.
[1]Corresponding author: pm389@cornell.edu

to 60% in untreated cows down to 0 to 15% in treated cows (Halasa et al., 2009) and DCT has also been associated with a decrease in clinical mastitis (**CM**) cases during lactation (Whist et al., 2006).

Although the majority of NIMI can be subclinical during the dry period, these infections can become clinical in early lactation (Green et al., 2002). It has been estimated that 55% of environmental infections established early in the dry period, including gram-negative IMI, can persist into the next lactation and can result in CM cases (Todhunter et al., 1995). In fact, 52% of all clinical coliform mastitis cases occurring in the first 100 DIM of lactation may originate during the previous dry period (Bradley and Green, 2000). Smith et al. (1985) also reported that the risk for NIMI from environmental pathogens can be 10 times higher during the dry period than during lactation.

Several authors have evaluated the efficacy between DCT and their association to both quarter- and cow-level outcomes during lactation (Arruda et al., 2013a,b; Scherpenzeel et al., 2014; Johnson et al., 2016) in herds in the United States. Although products undergo rigorous testing with a negative control before becoming publically available, no data is available for the Italian market comparing Cefovet A (**CF**; Merial Italia SpA, Milan, Italy), which contains cephazoline, and Cepravin (**CP**; MSD Animal Health srl, Segrate, Italy), which contains cephalonium dehydrate. These products are available in Italy and have a significant difference in both milk and meat withhold time, as well as pathogens on the label. There is a 0-h milk withhold after calving if the dry period length is at least 30 d for CF and 168-h (7 d) withhold with a 51-d dry period for CP. Both products are labeled to reduce the frequency of existing infections and limit new infections with *Streptococcus agalactiae*, *Streptococcus uberis*, *Streptococcus dysgalactiae*, and *Staphylococcus aureus*; however, CP also includes *Actinomyces pyogenes*, *Corynebacterium ulcerans*, *Escherichia coli*, *Proteus* spp., *Klebsiella* spp., *Citrobacter* spp., and *Enterobacter* spp.

The objective of the current study was to compare the efficacy of these 2 commercial DCT products in Italian herds in quarters also infused with an internal teat sealant. Efficacy was measured by quarter-level risk difference for cure of an IMI during the dry period, odds of a cure and development of NIMI over the dry period, and risk for experiencing a CM event between calving and 60 DIM. The hypothesis tested was that quarters infused with CF would have a noninferior proportion (a priori set delta = 0.1) of quarters cured from preexisting IMI, and no difference in the odds of a NIMI postcalving or risk of CM from calving to 60 DIM compared with quarters infused with CP.

MATERIALS AND METHODS

Study Design and Product Information

A randomized clinical field trial to evaluate noninferiority between 2 DCT products was conducted from March 2014 to November 2014 in 8 commercial dairy herds (A–H) in Italy. Cow enrollment began in March and continued until the predetermined sample size was enrolled; cows were then followed forward in time until 60 DIM. Eligible cows were randomly allocated to treat all 4 quarters with 1 of the 2 DCT according to a previously prepared randomized spreadsheet created in Excel software (Microsoft Corp., Redmond, WA). Randomization was blocked within farms. The authors residing at University of Milano and Istituto Zooprofilattico Sperimentale visited the herds weekly and conducted all study enrollment and collected milk samples at the 3 different time points [dry off (**S1**), within 2 to 9 DIM (**S2**), and within 10 to 17 DIM (**S3**)]. Milk from cows with CM up to 60 DIM was collected by trained on-farm personnel within each herd, with training and supervision conducted by the principal investigators.

Cefovet A contains 250 mg of cephazoline and 85 mg of aluminum distearate in a stable olive oil to reach a weight of 3 g. It is labeled for treatment and prevention of mastitis caused by *Strep. agalactiae*, *Strep. uberis*, *Strep. dysgalactiae*, and *Staph. aureus*. Milk withholding times are 0 d after calving if the dry period length is at least 30 d (14 d if the dry period length is shorter); meat withholding time is 0 d, with the exception of the mammary gland, which cannot be used for human consumption. The second antibiotic, Cepravin, is composed of 250 mg of cephalonium dehydrate, aluminum distearate, and liquid paraffin to reach a weight of 3 g. It is labeled to reduce the frequency of existing infections and prevent new infections caused by *Strep. agalactiae*, *Strep. uberis*, *Strep. dysgalactiae*, *Staph. aureus*, *A. pyogenes*, *C. ulcerans*, *E. coli*, *Proteus* spp., *Klebsiella* spp., *Citrobacter* spp., and *Enterobacter* spp. Milk withholding times are 51 d postinfusion plus 168 h (7 d) postcalving, whereas meat withholding time is 2 d.

Herd Selection

Herds were considered for inclusion in the study if they agreed to comply with the study protocol and had regular DHIA testing. This convenience sample of herds averaged 450 lactating cows (varying from 120 to 1,198), with a 1-yr average bulk tank SCC of 240 $\times$ 10^3 cells/mL (varying from 180 to 350 $\times$ 10^3 cells/mL), and a daily herd milk production average of 32