### ARTICLE IN PRESS



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# Effects of antibiotic dry-cow therapy and internal teat sealant on milk somatic cell counts and clinical and subclinical mastitis in early lactation

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#### ABSTRACT

The objective of this study was to determine the efficacy of an internal teat sealant (TS; Teatseal; Zoetis Australia, Silverwater, NSW, Australia), when used in combination with antibiotic dry-cow therapy (ADCT) administered at dry-off, on milk individual somatic cell count (ISCC), milk production and components, and the incidence of clinical and subclinical mastitis in dairy cows up to 60 d after calving, when compared with ADCT only. Multiparous Holstein, Jersey, or Holstein cross cows (n = 2,200) from 8 farms in southern and eastern Australia were randomly assigned to treatment of all 4 quarters with ADCT alone or with ADCT plus TS (ADCT + TS) at dry-off in this randomized, multisite clinical trial. Individual milk vield, fat and protein percentages, and ISCC were measured at intervals of  $14 \pm 3$  d after calving for the first 60 d of lactation. The first measurement occurred between 10 and 24 d after calving. Clinical mastitis and health events were recorded from dry-off to 60 d of lactation. Milk samples were collected from first cases of clinical mastitis and subjected to bacteriology. Treatment and the interaction of treatment by time did not affect milk yield, ISCC weighted by milk yield, or fat and protein percentages. Treatment with ADCT + TS decreased geometric mean ISCC compared with treatment with ADCT alone over the first 60 d of lactation. Geometric mean ISCC ( $\times 10^3$  cells/mL) was 32.0 [95% confidence interval (CI): 26.8 to 38.3] and 43.5 (95% CI: 36.2 to 52.1) for ADCT + TS and ADCT alone, respectively. The odds of at least 1 case of subclinical mastitis (ISCC  $\geq$ 250,000 cells/mL) were 1.9 times higher (95% CI: 1.4 to 2.6) with ADCT alone in the first 60 d of lactation compared with ADCT + TS. Use of ADCT + TSreduced the estimated incidence of at least 1 case of subclinical mastitis on all 8 farms, compared with use of ADCT alone. Only 4 cows that calved 40 to 100 d

after dry-off had a first case of clinical mastitis in the dry period. Five percent of cows (76 cases from 1,528) cows included in this analysis) that calved 40 to 100 d after dry-off had a first case of clinical mastitis between 0 and 60 d in milk. Of these first cases of clinical mastitis, 43 cases (5.7% of 761 cows) occurred in the ADCT group and 33 (4.3% of 767 cows) in the ADCT + TS group, but this was not significantly different. Proportional hazards estimates of survival showed no difference in the number of days postcalving to detection of first cases of clinical mastitis between the ADCT and ADCT + TS groups over the first 60 d postpartum. The estimated hazard ratio for clinical mastitis over this period in the ADCT + TS cows (relative to ADCT) alone) was 0.70 (95% CI: 0.43 to 1.14). The combination of ADCT and TS provides benefits over ADCT use alone through improved prevention of subclinical mastitis and reduced ISCC in the first 60 d of lactation. **Key words:** dry-cow therapy, individual somatic cell count, intramammary infection, mastitis, teat sealant

#### INTRODUCTION

Interventions that provide a means to reduce the risk of infection of clinical or subclinical mastitis and the sequelae to infection, such as elevated individual cow SCC (**ISCC**), independent of causal organisms, are of great potential benefit to the dairy industry (Rabiee and Lean, 2013). Antibiotic dry-cow therapy (ADCT) is widely used at the end of lactation. It is designed to cure existing IMI and prevent new IMI during the dry period (Smith et al., 1966). Despite the use of ADCT, clinical mastitis caused by environmental pathogens remains common. Bradley and Green (2000) showed that 52% of clinical coliform mastitis cases in the first 100 d of lactation occurred in guarters that were infected during the dry period. Most antibiotic formulations persist during the early to middle dry period but do not cover the entire dry period (Gruet et al., 2001).

Part of the cow's natural defense mechanisms against IMI after dry-off is the formation of a keratin plug in the teat canal that acts as a physical barrier to mastitis-

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causing pathogens. Failure or delay in formation of the keratin plug during the dry period is an important risk factor for new IMI. Dingwell et al. (2003) reported that quarters with "open" teat-ends and quarters that had cracked teat-ends were both 1.7 times more likely to develop new IMI during the dry period, compared with quarters that "closed" and that were not cracked. Williamson et al. (1995) showed that 45 to 55% of quarters had open teat-ends 7 d after dry-off, and 97% of the clinical infections in the 21 d after dry-off occurred in these open quarters. A combination of the benefits of ADCT and the use of an internal teat sealant  $(\mathbf{TS})$  to mimic the protective effects of the keratin plug to provide protection during the entire dry period is used in Australia, North America, Europe, and New Zealand. One such TS is Teatseal (Zoetis Australia, Silverwater, NSW, Australia), a nonantibiotic viscous formulation that is primarily composed of 65% bismuth subnitrate (inert salt) and liquid paraffin administered in a syringe similar to that of ACDT. The TS acts as a physical barrier to invasion of the teat canal by mastitis-causing pathogens for up to 100 d (Woolford et al., 1998).

A meta-analysis from Rabiee and Lean (2013) showed that bismuth subnitrate-based TS (Teatseal or Orbeseal; Pfizer Animal Health, West Ryde, Australia) in the presence of ADCT reduced the risk of clinical mastitis after calving in lactating cows by 48% [risk ratio ( $\mathbf{RR}$ ) = 0.52; 95% CI: 0.37 to 0.75].

The few studies on the effects of the combination of ADCT and TS on the estimated linear somatic cell score (LS) of milk ISCC after calving had differing responses. Godden et al. (2003) and Runciman et al. (2010) showed a reduced LS in cows treated with ADCT and TS compared with ADCT alone. Only a trend toward a reduction in LS was reported by Baillargeon and LeBlanc (2010) at the second herd test, but no treatment effects were found at the first or third herd tests. Mütze et al. (2012) found no difference in ISCC between ADCT and ADCT + TS treated cattle over the first 3 mo of lactation. Rabiee and Lean (2013) pooled raw data from Cook et al. (2005), Sanford et al. (2006), and Baillargeon and LeBlanc (2010), and showed that the estimated LS of cows treated with a combination of ADCT and TS was not significantly different from that of those treated with ADCT only; however, Rabiee and Lean (2013) concluded that further studies were needed in this area.

Studies on the effects of the combination of ADCT and TS on subclinical mastitis are scarce, but Runciman et al. (2010) found that the RR of subclinical mastitis, defined as ISCC  $\geq 250,000$  cells/mL at the first herd test (conducted 7 to 50 d after calving) in ADCT + TS cows compared with ADCT only was 0.80 (95%) CI: 0.65 to 0.98; P = 0.035), indicating a lower risk of subclinical mastitis in ADCT + TS cows.

The objective of this study was to determine the efficacy of a TS product (Teatseal; Zoetis Australia) when used in combination with ADCT administered at dryoff, on milk ISCC, milk production and components, and the incidence of clinical and subclinical mastitis of multiparous dairy cows up to 60 d after calving compared with ADCT only.

#### MATERIALS AND METHODS

#### Experimental Design

Multiparous cows from 8 commercial pasture-based dairy herds in southern and eastern Australia were enrolled in this multicenter, randomized controlled trial with blocking between May 2014 and June 2015. Each cow, provided she met the required study inclusion criteria, was enrolled from dry-off until 60 DIM. All experimental procedures were approved by the Scibus Animal Ethics Committee (Scibus 0414-1215) and complied with Australian Animal Welfare Regulations.

#### Herd Selection

Herds were selected on (1) location, (2) willingness to comply with the study protocol, (3) number of cows, (4) good biographical records of cows, (5) absence of *Mycoplasma bovis* on a bulk-vat PCR test (RtMastitis major-4, Dairy Technical Services Ltd., Kensington, VIC, Australia), and (6) eligibility of cows. Herds were not selected based on previous mastitis incidence or ISCC.

The target enrollment was 1,000 cows per treatment group. Sample size was determined based on a reduction in LS of 0.5 between an anticipated LS of 4.7 (approximately 30,000 in ISCC) with statistical power 0.9 at  $\alpha$ = 0.05, based on a single prior ISCC and 3 repeated samples using Stata (version 12.0; StataCorp LP., College Station, TX). The estimates used for the sample size calculation were derived from previous studies on TS and ISCC (Rabiee and Lean, 2013).

#### **Cow Selection**

Cows were eligible for enrollment if they (1) had no clinical signs of disease, (2) had a predicted dry period between 40 and 80 d, (3) were entering their second or greater lactation, (4) had 4 functioning quarters free from teat abnormalities with teat-end scores of  $\leq 3$  on a scale of 1 to 5 (Britt and Farnsworth, 2011), (5) lameness score  $\leq 2$  on a scale of 1 to 5 (Sprecher et al.,

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