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Effects of supplementation of active dried yeast and malate during sub-acute ruminal acidosis on rumen fermentation, microbial population, selected blood metabolites, and milk production in dairy cows

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ABSTRACT

The objective of this study was to evaluate the effects of dietary supplementation of malate (MAL) and active dried yeast (ADY) on feed intake, rumen fermentation parameters, rumen microbial populations, selected blood metabolites, and milk production during a sub-acute ruminal acidosis (SARA) challenge in primiparous lactating dairy cows. Six rumen-fistulated Holstein dairy cows (body weight: 630 ± 55 kg, 110 ± 25 days in milk, mean ± SD) were assigned to the following treatments in a 3 × 3 Latin square design: (1) control TMR (CON); (2) a TMR supplemented with 80 g of sodium–calcium malate/head per day (MAL); and (3) a TMR supplemented with 10 g of active dried yeast providing 20×10^9 CFU of *Saccharomyces cerevisiae*/head per day (ADY). Each experimental period consisted of 14 days of adaptation to the experimental treatments, 4 days of SARA challenge, and 10 days of rest. Dry matter intake (18.4 *vs.* 19.8 kg/day), and milk yield (29.3 *vs.* 30.4 kg/day) were depressed during SARA compared with adaptation. Malate and ADY had no effect on DMI and milk yield during the adaptation and SARA phases. Malate and ADY had no effect on ruminal pH characteristics during adaptation. During SARA, maximum and mean ruminal pH was not affected by supplementation, but minimum ruminal pH tended to be higher for ADY compared to CON and MAL. Time spent with ruminal pH <5.6 and < 5.8 tended be lower for ADY compared to CON. During adaptation, ADY tended to decrease rumen NH_3-N concentration compared to CON. Malate and ADY had no effect on selected blood metabolites, but glucose and insulin concentration increased during SARA compared to adaptation. The population of *Fibrobacter succinogenes* tended to increase with MAL and ADY during adaptation, but its population was decreased during SARA compare with adaptation. During SARA, the abundance of *Megasphaera elsdenii* tended to be higher for ADY than for MAL. The abundance of *Streptococcus bovis* increased during SARA, but was not affected by MAL or ADY supplementation. The relative abundances of protozoa in the rumen decreased during SARA. In conclusion, ADY supplementation to dairy cows with SARA can potentially improve rumen function, as indicated by a tendency for an improved ruminal pH and greater abundance of *M. elsdenii* within the rumen.

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However, supplementation of MAL provided no benefit to dairy cows under SARA condition, at least with the inclusion rates used in this study.

1. Introduction

In Iran and other regions with a low availability of high-quality forages, high-producing dairy cows are fed diets high in grains that are rich in rapidly fermentable carbohydrates (>40% NFC and NDF <32%, % of DM) to meet their energy requirements. When feeding such high-grain diets, the rate at which organic acids are produced by microbial fermentation can exceed the rate at which these acids are absorbed across the ruminal epithelium, leading to their accumulation within the rumen, thereby decreasing ruminal pH and predisposing cows to sub-acute ruminal acidosis (SARA) (Owens et al., 1998). Based on a continuous recording of ruminal pH, SARA was defined as a depression of ruminal pH below 5.6 exceeding 3 h per day (AlZahal et al., 2007). An Iranian field survey indicated that the incidence of SARA was 27% in early- and mid-lactation cows (Tajik et al., 2009). Penner et al. (2007) reported that primiparous cows are particularly susceptible to developing ruminal acidosis after parturition, because primiparous cows have not experienced a long-term exposure to a highly fermentable lactation diet. Additionally, Enemark et al. (2004) concluded that generally, primiparous cows were more prone to low ruminal pH, higher ruminal VFA concentrations, and possibly to metabolic acidosis than multiparous cows.

Subacute ruminal acidosis accounts for substantial economic losses in the dairy industry due to decreased feed intake, reduced milk production, milk fat depression, diarrhea, and laminitis (Owens et al., 1998). Several potential dietary strategies to reduce the incidence of SARA have been reported, including feeding unprocessed grains that are less-rapidly fermentable, providing TMR instead of component feeding, and feeding smaller and more frequent meals throughout the day (Krause and Oetzel 2005). I addition, Gonzalez et al. (2012) proposed that use of feed additives such as buffers, natural plant extracts, ionophores, yeasts or organic acids would be helpful to reduce the incidence of SARA.

Yeast products are commonly used for inclusion in diets of dairy cows (Poppy et al., 2012). Differences exist between active dried yeast (ADY) and yeast cultures. Yeast cultures that are produced through yeast fermentation contain fermentation by-products and are not dependent on live yeast for their physiological effects (Callaway and Martin, 1997). In contrast, ADY products are products that, by definition, must contain >15 billion live yeast cells/g. It was reported that *Saccharomyces cerevisiae* stimulate lactate utilization by *Megasphaera elsdenii* and *Selenomonas ruminantium* spp. *lactilytica* (Chaucheyras et al., 1996). The reduction in lactic acid concentration should increase ruminal pH and favor the growth of cellulolytic bacteria and thus fiber digestion (Lila et al., 2004). However, Vyas et al. (2014) concluded that yeast supplementation did not influence ruminal pH during a severe acidosis challenge as the efficacy of both viable and killed yeast was reduced at low ruminal pH in beef heifers.

In addition, Calsamiglia et al. (2012) suggested organic acids may play a role similar to ADY. Nisbet and Martin, (1990) observed that lactate uptake by *S. ruminantium* increased over 4-fold with aspartate and fumarate, and increased over 10-fold with malate (MAL). Studies that examined the effects of MAL supplementation on ruminal fermentation characteristics are scarce. It was reported that aspartate, fumarate, and MAL stimulated the growth of *S. ruminantium* because they overcome the deficiency of oxaloacetate associated with gluconeogenesis, either by metabolizing fumarate and MAL into oxaloacetate or by the sparing effect of aspartate (Castillo et al., 2004). Montaño et al. (1999) observed that supplementation of a high-grain finishing diets with malic acid (80 g per animal per day) promoted a higher ruminal pH. In another study, Carro and Ranilla (2003) evaluated the effects of different concentrations of disodium–calcium MAL on *in vitro* ruminal fermentation with corn, barley, wheat and sorghum as substrates. For all substrates, the final pH increased as MAL concentration increased.

Despite the evidence of ADY and organic acids altering rumen fermentation, to our knowledge, no research exists that compared the influence of ADY and MAL on mitigating SARA by utilizing a nutritional SARA induction model, and concurrently assessed changes in ruminal pH patterns and in ruminal microbial populations in mid-lactating cows. Therefore, the objective of this experiment was to evaluate the effects of ADY and sodium MAL supplementation on ruminal pH, rumen fermentation variables, microbial community, and cow performance (DMI, milk yield, and milk composition) during a dietary regimen that leads to SARA.

2. Material and methods

2.1. Animals, feeding, and sampling procedure

The experiment was conducted at the Research Farm of the Agriculture Faculty of Ferdowsi University of Mashhad. All animals were cared for according to the guidelines of the (Iranian Council of Animal Care, 1995). Six primiparous Holstein dairy cows (BW = 630 ± 55 kg, 110 ± 25 DIM, mean ± SD) fitted with a rumen cannula (10 cm; Bar Diamond Inc., Parma, ID) were used in a replicated 3 × 3 Latin square design with 28-day periods. Cows were fed 1 of 3 dietary treatments: (1) control TMR (CON); (2) a TMR supplemented with 80 g of a sodium–calcium MAL/head per d (MAL; Rumalato®; Norel & Nature Nutrition, Madrid, Spain); and (3) a TMR supplemented with 10 g of a yeast culture, providing 20×10^9 CFU of *S. cerevisiae*/head per day (ADY; Yea-Sacc®1026; Alltech, Nicholasville, KY, USA). Levels of MAL and ADY were based on