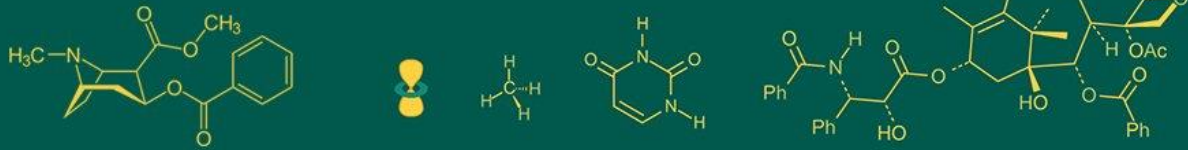


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## A comprehensive review on sub-acute ruminal acidosis in dairy cow

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### Abstract

Ruminal microbiota is pivotal to dairy cattle's digestive health, influencing ruminal pH and, in turn, the development of metabolic disorders. Subacute ruminal acidosis (SARA) arises from an imbalance between lactic acid-producing and lactic acid-consuming bacteria. Factors such as alterations in volatile fatty acid (VFA) absorption rates, gene expression, intracellular pH regulation by ruminal cells, and in situ carbohydrate digestibility play pivotal roles in elevating the risk of SARA in dairy cows. In SARA cases, ruminal pH levels drop to 5.5-5.0, promoting conditions like laminitis, impacting body condition scores, and increasing the incidence of liver abscesses. The threat of SARA can be mitigated through improved feeding strategies and rigorous feed management practices. Effective feeding strategies must strike a delicate balance between ruminal buffering and the production of volatile fatty acids through carbohydrate fermentation. Implementing total mixed ration (TMR) feeding has shown promise in reducing SARA occurrences. This comprehensive review delves into the prevalence, underlying causes, diagnostic methods, preventive measures, and the management and treatment options available for Subacute Ruminal Acidosis in dairy cattle.

**Keywords:** Dairy cow, Laminitis, Ruminal pH, SARA

### Introduction

The production of dairy cows has improved dramatically throughout the previous few decades. Nutrient requirements increased because of this development. To accomplish it, farmers fed cows with high grains diet in large amount which affect the health of cow (Sundrum, 2015) [68]. This type of feeding strategies can cause metabolic disorder followed by affecting the ruminal pH and indirectly on the health of animals. In SARA, ruminal pH is decreased for few hours per day up to pH= 5-5.5 because of VFA accumulation and inadequate buffering action of rumen. The survey reported that incidence of SARA is between 19% and 26% in early and mid-lactation respectively, in dairy cows. According to Bramley *et al.* (2008) [7] and Aschenbach *et al.* (2011) [3], SARA generally resulted from diets low in digestible fiber or rich in simply fermentable carbohydrates. The dairy cows with SARA do not show any typical clinical signs (Mutsvangwa *et al.*, 2002; Krause and Oetzel, 2005; Tajik and Nazifi, 2009) [52, 41, 69]. In lactating cow, SARA is also related to inflammation of various organs and tissues. The cows with SARA exhibited decreased rumen motility, loss of body condition score (BCS), diarrhea, depression, reduced milk production, inflammation, liver abscesses in and laminitis (Nocek, 1997; Stone, 2004; Alzahal *et al.*, 2007) [53, 67, 1]. It is believed that SARA may affect fermentation process due to excess VFA concentration and increase ratio of acetic acid to propionic acid that shifted towards propionic and butyric acids (Nordlund and Garrett, 1994) [55]. Further studies are necessary to know the function or role of valeric acid in developing SARA, which might be the potential marker to detect SARA (Morgante *et al.* 2007) [51]. The difference between acid production and acid removal from the rumen determines the pH of the rumen.

Acid can be eliminated in three ways: absorption by the ruminal epithelium, buffer neutralization, and transit from the rumen to the lower intestine. It has been implicit that the saliva production is increased by extending the spent chewing duration, *i.e.*, eating and ruminating. Saliva has inorganic buffers, such as sodium bicarbonate, which help to neutralize the organic acid formed during rumen fermentation (Church, 1988) [12].

By comparing healthy control cows with SARA affected cows, higher concentrations of short-chain fatty acids (SCFA) found in rumen along with lower protein expression and gene expression of SLC5A8 in rumen epithelium of SARA cows lower in SARA cows (Zhao *et al.*, 2020) [75]. To reduce the occurrence of SARA, the NRC (2001) [56] suggests a maximum feeding of non-fibre carbohydrates (NFC) and a minimum level of NDF. To overcome this delinquency, a new concept of peNDF (physically effective fibre) has been established which imitates feed ability for stimulation of salivation buffer and chewing in the ruminal environment (Mertens, 1997) [48]. Stone (1999) [66] has studied in 500 –dairy cow in New York and calculated US\$ 400 to US\$ 475 lost income per cow per year due to SARA. This estimate was based on various production related parameters observed like reduction in milk output of 3 kg/animal/day, reduced milk fat and true protein 34 g/kg and 29 to 28 g/kg, respectively. However, cost of other disorder which has an impact on reproductive organs was not estimated.

**Prevalence**

Accurately determining the prevalence of Subacute Ruminal Acidosis (SARA) can be challenging due to the difficulty in diagnosing the condition. Nonetheless, various studies have shed light on its occurrence in different regions.

The Khorasan Razavi province of northeast Iran, 196 rumen fluid samples were collected from ten dairy herds through rumenocentesis. It was discovered that 54 out of 196 cows (27.6%) were diagnosed with SARA (Tajik *et al.*, 2009) [69]. In Greek dairy herds, SARA-positive herds were identified in 33% of cases, while SARA-marginal cows were present in 8.33% of herds, with 58.33% being SARA-negative (Kitkas *et al.*, 2012) [37]. A cross-sectional study conducted in the central region of the UK found that 26.2% of 244 primarily early lactation dairy cows exhibited signs of SARA (Atkinson, 2014) [5]. In Polish dairy farms, 14% of cows had pH levels below 5.6, and SARA was detected in 44% of the herds, potentially due to increased starch utilization (Stefanska *et al.*, 2016) [65]. An experiment involving 120 cows found that 10.83% had pH levels of 5.5 or higher, with some cows falling into the marginal category (6.6% at pH 5.6-5.8), while the majority were considered normal (82.5% at pH>5.8) (Ural *et al.*, 2017) [17]. Research conducted in Dutch dairy farms reported an overall prevalence rate of 13.8% for SARA, with farm-specific rates ranging from 0% on some farms to as high as 38% on others (Kleen *et al.*, 2013) [32]. These findings collectively highlight the variable prevalence of SARA in different geographic regions and underscore the importance of monitoring and managing this condition in dairy cattle.

**Table 1:** Studies on ruminal acidosis in dairy a cow

Sr. No.	Study	Prevalence rate	References
1.	A survey of 15 dairy farms in Wisconsin in lactating cows	19% in early lactation cows 26% in mid lactation cows	Garrett <i>et al.</i> (1997) [25]
2.	A survey on 14 dairy farms in Wisconsin in lactating cows	20.1% of early and peak lactation cows	Oetzel <i>et al.</i> (1999) [58]
3.	Subacute ruminal acidosis (SARA) in grazing Irish dairy cows	11%	O'Grady <i>et al.</i> (2008) [59]

**Etiology of SARA**

The etiology of Subacute Ruminal Acidosis (SARA) encompasses several contributing factors, primarily rooted in nutrition. Key causes include:

**High Concentrate Diet and Low Crude Fiber:** Diets rich in concentrates and deficient in crude fiber can predispose cattle to SARA.

**Inadequate Forage Particle Length**

When forage particles are too short, it can disrupt the normal digestive processes.

**Limited Buffering Mechanisms:** Insufficient buffering capacity in the rumen can exacerbate acidosis.

This nutritional imbalance can lead to disruptions in the balance between lactic acid-producing and lactic acid-utilizing bacteria. As a result:

Lactic acid can accumulate in the rumen, potentially triggering conditions like laminitis. There is an increase in volatile fatty acid (VFA) production, further lowering the rumen pH.

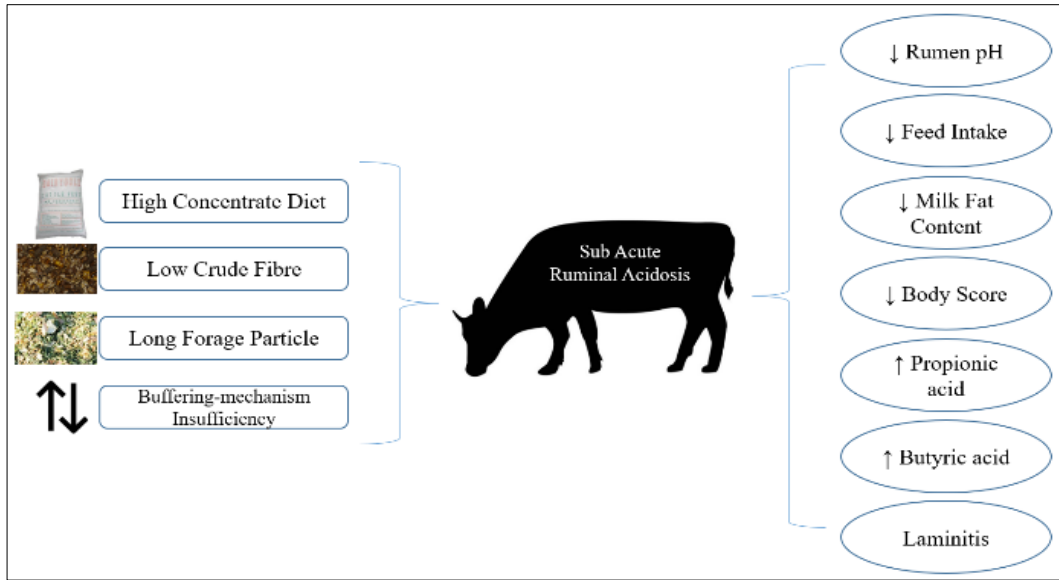
In addition to the production and accumulation of VFAs and protons, VFA absorption plays a crucial role, involving the exchange of ions like Na<sup>+</sup> and HCO<sup>3-</sup> within the epithelial tissue. This ion exchange helps regulate intracellular pH and VFA absorption (Aschenbach *et al.*, 2011; Penner *et al.*, 2011) [3, 62].

Furthermore, rumen pH is influenced by the production of acids during the fermentation of organic materials (Chen *et al.*, 2012) [10].

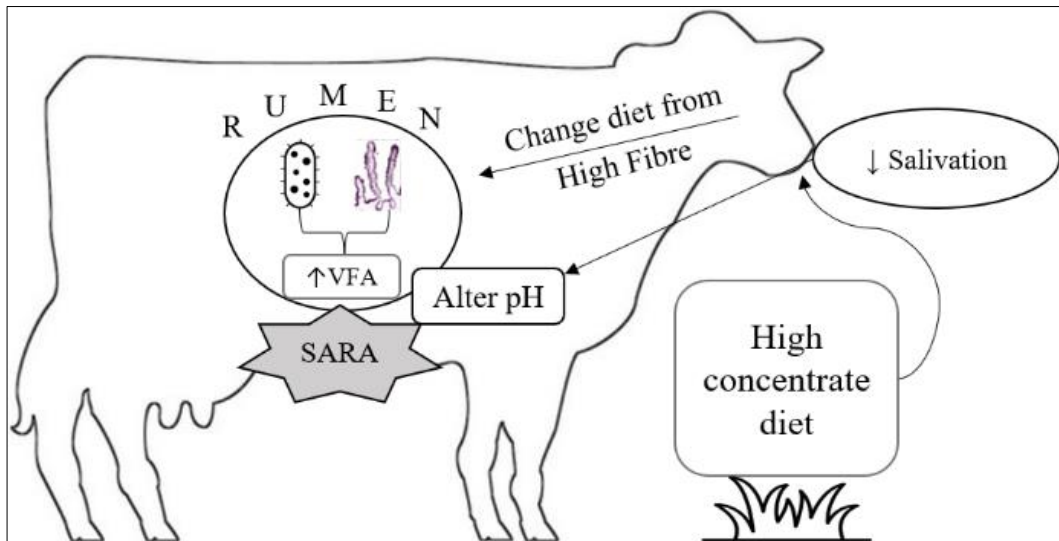
It is noteworthy that extended chewing time, encompassing both eating and ruminating, stimulates saliva production. Saliva contains inorganic buffers, such as sodium bicarbonate, which aid in neutralizing the organic acids generated during rumen fermentation (Church, 1988) [12].

**Table 2:** Causes and sequel of a dairy cattle

Sr. No.	Studies	Causes	Reference
1.	Clinical Aspects of Ruminal Acidosis in Dairy Cattle	1. Excessive intake of rapidly fermentable carbohydrates 2. Inadequate ruminal buffering 3. Inadequate Adaptation to Highly Fermentable, High Carbohydrate Diets	Oetzel (2000)



**Fig 1:** Development of SARA in cow



**Fig 2:** Consequences of SARA in dairy cow

Figure 2. Development of SARA in cow. It occurs when abrupt change in diet from high fibre to high concentrate occurs leading to difficulty in ruminal bacteria and papillae adjustment turning into rapid and increased production of VFA producing SARA. Also, reduction of fibre content in diet suppresses rumination and hence salivation which changes the ruminal pH leading to production of SARA.

**Effect on rumen and hindgut**

Sudden transitions in diet, shifting from a high-fiber to a high-concentrate diet, can precipitate Subacute Ruminant Acidosis (SARA). This abrupt change challenges the adaptation of ruminal bacteria and papillae, resulting in the rapid production of volatile fatty acids (VFAs), a hallmark of SARA. Additionally, reducing dietary fiber content can suppress rumination and salivation, further disrupting ruminal pH and promoting SARA.

In SARA cases, there is an accumulation of short-chain fatty acids (SCFAs) in the rumen, leading to a decrease in ruminal pH and alterations in the rumen microbiota. These shifts result in reduced fiber and total carbohydrate digestion, leading to energy loss. Occasionally, cows may exhibit reduced body condition without a corresponding

decrease in feed consumption (Hall, 2002; Kleen *et al.*, 2013; Dijkstra *et al.*, 2012) [30, 32, 18].

Transitioning beef heifers from a primarily grass hay diet to one dominated by barley grain can induce changes in the bacterial communities attached to the rumen epithelium. During heavy grain feeding, species such as *Ruminobacter*, *Treponema*, and *Lachnospiraceae* become prominent (Chen *et al.*, 2011) [11]. Changes in rumen bacterial populations are considered an initial step in SARA's detrimental effects on animal performance. During SARA, potentially harmful and inflammatory substances, including endotoxin (lipopolysaccharide or LPS), increase alongside shifts in microbial populations.

Fermentable carbohydrates in a SARA-inducing diet can lead to exponential bacterial growth, followed by significant bacterial lysis due to decreasing substrate availability, reduced rumen pH, and the accumulation of fermentation byproducts (Zebeli and Metzler-Zebeli, 2012) [74].

The rumen epithelium serves as a selective barrier, allowing the absorption of SCFAs while preventing bacterial entry and colonization.

A disruption in this barrier is necessary for SARA to have systemic consequences. The four layers comprising the

rumen epithelium stratum corneum, stratum granulosum, stratum spinosum, and stratum basale play distinct roles. In a healthy rumen, bacteria are loosely associated with the stratum corneum. The stratum granulosum and, to a lesser extent, the stratum spinosum express tight junction proteins that govern permeability.

SCFAs can be transported from rumen contents to the basal lamina, establishing connections between the stratum granulosum, stratum spinosum, and stratum basale. The rumen's permeability barrier adapts to changes, such as oxidative stress or thermal stress possibly as an adaptive response to higher grain diets, allowing for increased SCFA absorption (Zebeli and Metzler-Zebeli, 2012) [74].

Furthermore, research has reported increased permeability in response to acidification or hyperosmolality in isolated rumen sections (Aschenbach and Gabel, 2000; Emmanuel *et al.*, 2007; Penner *et al.*, 2010) [3, 21, 62].

During SARA, changes in the rumen are mirrored in the large intestine. Increased SCFA and LPS levels, along with decreased pH and intestinal mucosa damage, coincide with heightened carbohydrate fermentation, contributing to SARA (Dijkstra *et al.*, 2012; Li *et al.*, 2012) [18, 42, 43].

Faecal indicators of SARA may include diarrhea, frothy feces, larger particle size in feces, and the presence of mucin casts (Hall, 2002) [30]. The systemic inflammatory effects of SARA have been suggested to result from germs or toxins passing through the intestinal mucosa, as the intestinal epithelium consists of a single layer of epithelial cells. In fact, found that the presence of LPS in the bloodstream following a SARA challenge indicated that LPS entered the circulation via the intestines rather than the rumen.

### Decreased feed intake

An increase in rumen fluid osmolality exceeding 300 mOsm/L in cows affected by Subacute Ruminal Acidosis (SARA) can lead to limitations in feed intake and disrupt bacterial fermentation of fiber and starch (Carter and Grovum, 1990) [9]. Reduced dry matter intake (DMI) is considered a consistent clinical indicator of SARA (Garry, 2002) [27]. Several studies have demonstrated that lower feed intake during SARA episodes can be beneficial (Brown *et al.*, 2000; Krajcarski-Hunt *et al.*, 2002) [8, 40].

In certain studies, it was observed that cows altered their diet preferences to mitigate SARA, such as consuming more hay when given the option. Interestingly, when provided with choices, cows did not select sodium bicarbonate.

Grain-induced SARA has been shown in several studies to result in an increase in acute phase proteins in the bloodstream, indicative of inflammation (Gozho *et al.*, 2005, 2006, 2007) [28, 29]. Inflammation in various organs can lead to reduced feed intake in cows (Weingarten, 1996; Andersen *et al.*, 2000) [72, 2]. Consequently, the inflammation associated with grain-induced SARA may play a role in the reduction of feed intake. This hypothesis gains support from studies showing that inducing SARA by substituting alfalfa hay with alfalfa pellets did not result in inflammation or a decrease in feed intake (Khafipoor *et al.*, 2007) [34].

### Milk composition

The addition of buffering substances to a high-concentrate diet effectively restored a higher ruminal pH and prevented a decline in milk-fat content, as demonstrated by Khorasani and Kennelly in 2001. Multiple studies have suggested that the varying response of milk fat levels in experimentally

induced Subacute Ruminal Acidosis (SARA) may be correlated with the duration of SARA episodes. However, short-term SARA challenges have been shown to have no impact on milk composition (Krause and Oetzel, 2005) [41].

Research by Alzahal *et al.* (2010) [61] revealed that ruminal infusion of soybean oil (SBO) in combination with a moderate- or high-forage diet led to reduced ruminal pH and a more pronounced decline in milk fat production. SARA has also been associated with alterations in odd-chain and branched-chain fatty acids, possibly attributed to changes in rumen microbiota that can serve as indicators for detecting SARA. Milk fatty acid profiles have been proposed as potential indicators of acidosis occurrence, offering insights into its origin (Colman *et al.*, 2013) [14]. In a comparison with a control group, the SARA-induced group exhibited lower milk fat content, specifically at 4.14 percent (Danscher *et al.*, 2015) [16].

A limited number of studies have reported reduced milk fat content during SARA (Xu *et al.*, 2016; Bipin *et al.*, 2016; Malekkhahi *et al.*, 2016) [73, 6, 45]. Zschesche *et al.* (2020) [46] identified a significant correlation between the milk Fat: Protein ratio and ruminal pH parameters, which can be indicative of SARA in dairy cows. However, it's worth noting that not all experimental SARA simulations result in low milk fat content (Enjalbert *et al.*, 2008) [23]. Some authors have demonstrated that cows affected by SARA did not exhibit milk fat depression under farm conditions (Tajik *et al.*, 2009) [69]. In certain cases, SARA had no discernible influence on milk production or other milk components in high-producing cows across different stages of lactation (Kitkas *et al.*, 2019) [36]."

### Body condition Score

SARA in lactating cattle indeed has an impact on body condition scores. Ural *et al.* (2017) [17] found that cows suspected of having SARA typically exhibited body condition scores ranging from 2.75 to 3.75. It's worth noting that in herds affected by SARA, the presence of thin dairy cows with low body condition scores is a common occurrence, as reported by Kleen *et al.* (2003) [39]. However, it's important to highlight that the body condition score alone may not be a reliable tool for distinguishing between SARA-affected and unaffected dairy herds, as suggested by Kleen (2004) [38] and Tajik *et al.* (2009) [69].

### Metabolic Acidosis

In sub-acute ruminal acidosis (SARA), the relative proportion of propionic acid to butyric acid in the rumen increases, as observed in the study by Da-cheng *et al.* in 2013. Among the short-chain volatile fatty acids (VFAs), only acetic acid enters the peripheral circulation. However, butyric acid undergoes conversion primarily within the rumen wall into -hydroxy-butyric acid, potentially leading to a form of alimentary ketosis. In the liver, propionic acid is transformed into glucose, a process described by Owens *et al.* in 1998 [76] and Enemark in 2002 [22].

### Laminitis

It is also a frequent underlying factor contributing to hoof-related issues in cattle, such as sole ulcers, white line abscesses, and solar hemorrhages (Clarkson *et al.*, 1996; Smilie *et al.*, 1996; Warnick *et al.*, 2001) [13, 64, 71]. SARA can lead to damage to the ruminal epithelium, facilitating the absorption of histamine and endotoxins. These

substances, along with potentially others, disrupt the normal circulation and provoke inflammation within the hoof, ultimately leading to the development of laminitis (Vermunt, 1992) [70].

### Diagnosis

Kitkas *et al.* (2019) [36] demonstrated that the diagnosis of subacute ruminal acidosis (SARA) in dairy cows can be accomplished through various methods, including body condition score (BCS) assessment, clinical examination, rumen fluid sampling, and milk sampling. Notably, they reported no significant alterations in blood parameters during SARA, consistent with the findings of Hernández *et al.* (2014) [33]. Nevertheless, blood examinations may reveal signs of stress, such as leukocytosis with neutrophilia, as well as anemia and a decrease in packed cell volume due to ruminal ulcers and reduced appetite. In addition to BCS and clinical evaluation, Li *et al.* (2012) [43] suggested that SARA diagnosis can involve rumen pH measurement, rumen fluid sampling and analysis, feces and urine sampling and analysis, milk sampling, and blood sampling. These comprehensive methods collectively provide a robust approach to diagnosing SARA in dairy cows.

### Prevention

In numerous dairy farms, Subacute Ruminal Acidosis (SARA) is a condition with far-reaching consequences that not only impact the health of dairy cows but also have economic implications for the entire country. Fortunately, SARA can be mitigated through effective nutritional strategies. One such strategy involves the use of chemical buffers, a practice advocated by experts such as Hutjens (1991) [31] and Erdman (1988) [24]. Chemical buffers have been demonstrated to be effective in preventing acidosis in dairy cows, as noted by Garry (2002) [27]. These buffers can be seamlessly incorporated into feed rations, particularly when the dietary fiber content is insufficient to maintain rumen health. For instance, research has shown that supplementing feed with 150 grams of sodium bicarbonate daily resulted in increased milk production, a finding supported by Downer and Cummings (1985) [20]. By implementing such nutritional interventions, the adverse impacts of SARA on both cow health and the national economy can be mitigated.

#### a. SARA and direct fed microbials (DFM)

Yeast cultures have gained recognition as valuable additions to livestock feed rations. While many yeast-based products contain a mix of living and deceased yeast cells, research findings indicate that the inclusion of yeast can yield diverse effects, as documented by studies like those conducted by Aslan *et al.* (1995) [4].

Notably, Nocek and Kautz's (2006) [54] demonstrated that the introduction of 105 cfu/mL of three distinct microorganisms, namely *Lactobacillus plantarum*, *Enterococcus faecium*, and *Saccharomyces cerevisiae* led to a reduction in diurnal ruminal acidosis while enhancing the digestion of corn silage.

In another study, early lactation cows were provided with Direct-Fed Microbials (DFM) in the form of yeast and *E. faecium*, both supplemented at  $5 \times 10^9$  cfu/day. This intervention resulted in increased rumen digestion of dry matter in the fodder, higher milk production, and elevated dry matter consumption. However, it should be noted that

milk fat content was reduced, likely due to the stimulation of lactolytic flora, as reported by Martin and Dean (1989). They also explored genetic manipulation of lactolytic bacteria to enhance their lactate conversion ability and acid tolerance, although no commercially available product has been developed to date.

#### b) Carboxylic acid

The supplementation of dicarboxylic acids such as fumarate and maleate may also have similar effects, although concrete evidence supporting this notion is currently lacking, as outlined by Owens *et al.* (1998) [76].

#### c) Antibiotics

Antibiotics have been suggested as a means of preventing SARA and reducing lactate production by bacteria such as *S. bovis* and *Lactobacillus spp.* Additionally, ionophores like monensin have been shown to enhance total tract nutrient digestion without significantly affecting dry matter intake, milk production, composition, or ruminal pH when incorporated as a premix for lactating cows with grain-induced SARA, as demonstrated by Osborne *et al.* (2004) [77].

It's worth noting that monensin did not appear to influence rumen pH during SARA, as reported by Mutsvangwa *et al.* (2002) [52]. Consequently, the use of such chemical interventions may not always yield consistent results. Furthermore, it is important to acknowledge that the use of certain medications for maintaining ruminants on a non-ruminant diet, as discussed by Dirksen (1985) [19], remains a topic of debate, with varying perspectives on its necessity and effectiveness. Additionally, it is important to note that the use of antibiotics as a preventive measure should be done judiciously and in compliance with relevant regulations and guidelines.

d) Shalaan *et al.* (2023) [63] reported that ruminal juice supplemented with vitamins and minerals mix with ruminal anti-acid has a good impact on cattle clinically affected with ruminal acidosis.

#### Management tools

The quantity and timing of meals play a pivotal role in the dietary management of Subacute Ruminal Acidosis (SARA). Dairy cattle exhibit effective rumen pH self-regulation when they have consistent and predictable access to the same Total Mixed Ration (TMR) daily. Conversely, even minor feed restrictions can lead to overconsumption during meals. Milton (1998) [49] highlighted the significant impact of deviating from a regular feeding schedule by 2-4 hours, which significantly increased the risk of SARA in feedlot cattle. Hernandez-Urdaneta *et al.* (1976) [33] found that providing cows with a TMR instead of separate feeds helped avoid the consumption of large grain meals, thereby reducing the risk of acidosis.

Supporting these findings, Stergaard and Grohn (2000) [60] also observed that feeding concentrates separately from forage yielded better outcomes than adjusting the concentrate-to-forage ratio in a TMR, which was linked to a higher risk of metabolic disorders. When utilizing a TMR, there is greater control over the concentrate-to-forage ratio in the cow's diet. For instance, Maekawa *et al.* (2002) [44] discovered that cows fed a predetermined 50:50 ratio of concentrate to forage (on a dry matter basis) exhibited different ruminal pH dynamics compared to those fed a

TMR with a 50:50 concentrate-to-forage ratio. In particular, cows on the TMR diet showed a lower minimum ruminal pH, highlighting one advantage of TMR feeding in potentially preventing low ruminal pH caused by excessive concentrate intake.

**Table 3:** Difference between acute ruminal acidosis (ARA) and SARA

	Acute ruminal acidosis	Subacute ruminal acidosis
Rumen Parameters		
Rumen pH	Below 5	5-5.4
Volatile fatty acids	Decrease (<100mM)	Increase (150–225mM)
Gram negative bacteria	Decrease	Normal
Gram positive bacteria	Increases	Normal
Lactic acid producers	Increases	Increases
Lactic acid consumers	Decrease	Increase
Blood parameters		
Blood pH	Low	Borderline
Lactate	Increases	Normal
Bicarbonate	Low	Borderline

### Conclusions

Subacute Ruminal Acidosis (SARA) has a significant impact on both the well-being of dairy cows and the financial viability of dairy herds. The prevalence of SARA tends to rise as cows increase their total Dry Matter (DM) consumption, particularly when they are fed diets with higher proportions of grains. Unfortunately, this can lead to a potential decrease in milk production. It's essential to recognize that the short-term benefits of heavy grain feeding may be outweighed by the long-term consequences. In light of these challenges, dairy producers and nutritionists must work collaboratively to design diets that optimize energy intake and overall milk production while minimizing the risk of SARA. An integral aspect of SARA prevention is ensuring adequate ruminal buffering, which encompasses both dietary and endogenous buffering mechanisms. This can be achieved by incorporating physical fiber in the diet, which cannot be isolated from other feed components, and by maintaining an appropriate balance of cations and anions in the diet. Another preventive strategy against SARA involves restricting the intake of rapidly fermentable carbohydrates. This can be achieved by supplying sufficient dietary fiber, effectively processing grains, introducing high-fiber concentrates if necessary, and implementing a feeding regimen that allows cows to consume smaller, more frequent meals regularly. In addition to these dietary strategies, the use of feed additives and supplements can provide an additional layer of protection against the development of SARA. This comprehensive approach to SARA prevention ensures the welfare of dairy cows and sustains the profitability of dairy herds.

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