

Review

Ketosis as the Most Important Metabolic Disease in Dairy Cattle

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Abstract: Ketosis is a major metabolic disorder in dairy cattle, particularly during the transition period, with profound implications for health, productivity, reproduction, immunity, and welfare. This review synthesizes current knowledge on the pathophysiology, risk factors, detection strategies, interventions, and broader impacts of ketosis, highlighting recent advances and identifying research priorities. Transition cows experience dramatic metabolic and hormonal changes around parturition, with negative energy balance triggering excessive mobilization of adipose tissue and hepatic lipid accumulation. Body condition, abdominal adiposity, and energy reserves are critical predictors of metabolic risk, influencing susceptibility to both clinical and subclinical ketosis. Recent multi-omics studies, including metabolomics and lipidomics, have revealed complex biochemical signatures that enhance understanding of disease mechanisms and provide potential biomarkers for early detection. Parallel research on the rumen microbiome emphasizes the role of microbial–host interactions in modulating energy balance, volatile fatty acid production, and inflammatory responses. Dysbiosis and altered microbial metabolites can exacerbate metabolic stress, linking gut ecology directly to hepatic function and ketone body accumulation. Advances in precision monitoring and sensor integration now enable continuous tracking of behavioral, physiological, and production parameters, facilitating predictive identification of at-risk cows. Nutritional interventions, including controlled-energy diets, glucogenic precursors, rumen-protected choline, and methionine, remain foundational, while pharmaceutical approaches and microbiome-targeted strategies, such as probiotics, yeast cultures, and experimental fecal microbiota transplantation, offer complementary avenues for prevention and management. Ketosis exerts wider systemic effects, impairing reproductive performance, suppressing immune function, increasing antimicrobial use, and compromising welfare. These consequences underscore the need for integrated management strategies that align productivity, health, and sustainability objectives. Despite advances, knowledge gaps persist in longitudinal validation of biomarkers, mechanistic understanding of host–microbe interactions, and practical implementation of precision tools across diverse farm settings. Future research should prioritize multi-omics integration, individualized intervention strategies, and evaluation of economic and sustainability outcomes.

Keywords: *Subclinical ketosis; transition cow; metabolomics; lipidomics; rumen microbiome; precision dairy farming.*

1. Introduction

Ketosis has long been recognized as a critical metabolic disorder in dairy cattle, traditionally defined by elevated circulating ketone bodies, particularly β -hydroxybutyrate (BHB), during early lactation [1-4]. Historically, attention was focused primarily on clinical cases manifesting with reduced appetite, decreased milk yield, and observable lethargy. However, contemporary research has redefined ketosis as a multifaceted, systemic condition that encompasses subclinical manifestations, complex metabolic dysregulation, and extensive interactions with reproduction, immunity, the rumen microbiome, and overall animal welfare. In modern high-yielding dairy systems, where genetic selection, intensive feeding strategies, and precision management coalesce, this broader perspective is essential for understanding the true impact of ketosis and implementing effective control strategies [2]. The transition period, spanning the three weeks before and after calving, is now recognized as the most vulnerable window for metabolic disease. During this phase, cows face dramatic shifts in energy demand and endocrine status, with negative energy balance triggering excessive adipose tissue mobilization and hepatic lipid accumulation. Body condition scoring and the assessment of abdominal adiposity have emerged as practical tools to quantify metabolic risk, linking prepartum energy reserves with susceptibility to both subclinical and clinical ketosis. These phenotypic indicators, combined with detailed metabolic profiling, provide a foundation for individualized risk assessment and targeted intervention. Advances in multi-omics technologies, including metabolomics and lipidomics, have refined our understanding of the biochemical signatures underlying ketosis, identifying novel biomarkers and mechanistic pathways that extend beyond traditional measures. In parallel, rumen microbiome research has revealed that microbial–host interactions play a pivotal role in energy balance, volatile fatty acid production, and immune modulation, highlighting the gut as a critical contributor to metabolic resilience or vulnerability [3]. These insights underscore the complexity of ketosis as a disease shaped by both host physiology and microbial ecology. Emerging precision monitoring and sensor technologies are transforming the landscape of ketosis detection, enabling continuous measurement of feeding behavior, rumination, activity, milk composition, and physiological parameters. When integrated with machine learning algorithms, these tools allow early, predictive identification of at-risk cows, facilitating timely nutritional, pharmaceutical, and microbiome-targeted interventions. Nutritional strategies—including controlled-energy diets, glucogenic precursors, and rumen-protected nutrients—remain central, while pharmaceutical and microbial interventions complement these approaches, collectively aiming to restore metabolic balance and prevent disease progression. Beyond individual cow health, ketosis exerts wider impacts on reproduction, immunity, antimicrobial use, and welfare. Reduced conception rates, impaired immune responses, increased susceptibility to mastitis and metritis, and compromised welfare outcomes highlight the systemic nature of the disorder. Economic considerations and sustainability concerns further amplify the significance of effective ketosis management, as early detection and preventive strategies can reduce losses, minimize antimicrobial reliance, and enhance resource efficiency in modern dairy operations [1]. This review integrates contemporary knowledge across these domains, re-defining ketosis as a complex, systemic, and multifactorial condition in modern dairy systems. By examining transition physiology, metabolic profiling, microbial interactions, precision monitoring, interventions, and broader herd-level impacts, we aim to provide a comprehensive framework for understanding, detecting, and mitigating ketosis, ultimately supporting animal health, productivity, welfare, and sustainability in high-performing dairy herds [1-4].

2. Transition cow physiology and pathogenesis

The transition period in dairy cattle, extending from approximately three weeks before calving to three weeks after parturition, represents one of the most physiologically dynamic and vulnerable stages of the bovine life cycle [9-12]. It is during this time that the cow must successfully adapt from a pregnant, non-lactating state to one of intense milk synthesis, requiring major metabolic, endocrine, and immunological adjustments. Despite remarkable evolutionary adaptations that

enable high-yielding dairy cows to initiate lactation, these adjustments often create an environment of metabolic stress that predisposes animals to disorders such as ketosis. In recent years, advances in molecular biology, systems physiology, and precision monitoring technologies have significantly reshaped our understanding of the underlying mechanisms driving ketosis during the transition period. Central to the pathogenesis of ketosis is the negative energy balance (NEB) that inevitably accompanies the onset of lactation [8]. At calving, the sudden exponential increase in nutrient requirements for milk synthesis cannot be matched by dietary intake, as dry matter intake typically lags behind energy demand for several weeks postpartum. This mismatch compels the cow to mobilize endogenous body reserves, particularly adipose tissue, to provide non-esterified fatty acids (NEFA) as alternative energy substrates. The mobilized NEFA are taken up by the liver where they face three possible metabolic fates: complete oxidation through the tricarboxylic acid (TCA) cycle to generate adenosine triphosphate (ATP), partial oxidation to ketone bodies (primarily beta-hydroxybutyrate, BHBA), or re-esterification into triglycerides. Under conditions of high NEFA influx, the hepatic capacity for oxidation and very-low-density lipoprotein (VLDL) export becomes overwhelmed, leading to excessive ketogenesis and fat accumulation in the liver. This dual pathology of hyperketonemia and fatty liver has become recognized as the principal metabolic disturbance of transition cows. New insights from molecular studies have revealed that the regulatory mechanisms governing hepatic lipid metabolism are more complex than previously appreciated. Gene expression profiling and lipidomic analyses have identified key regulators such as carnitine palmitoyltransferase (CPT), acetyl-CoA acetyltransferase (ACAT), and microsomal triglyceride transfer protein (MTTP) as pivotal in balancing lipid oxidation and export. Furthermore, peroxisome proliferator-activated receptors (PPARs) have been implicated as master transcriptional regulators orchestrating the response of hepatocytes to NEFA overload. Evidence suggests that in cows with subclinical ketosis, downregulation of PPAR- α and impaired activation of oxidative pathways result in a metabolic bottleneck that shifts the fate of fatty acids toward ketogenesis and triglyceride storage rather than efficient energy production. These molecular insights have important implications for both prevention and therapeutic intervention, highlighting potential targets for nutritional modulation and pharmacological support. Another dimension of transition cow physiology that has gained attention is the role of gluconeogenesis and glucose partitioning. Glucose is a critical substrate during early lactation, not only as the primary precursor of lactose synthesis in the mammary gland but also as an energy source for immune cells and other vital tissues [11]. However, ruminants are entirely dependent on hepatic gluconeogenesis for glucose supply, as negligible amounts are absorbed directly from the gut. During NEB, increased demands for glucose cannot be met, leading to hypoglycemia and further reliance on ketone bodies as alternative fuels. Propionate derived from rumen fermentation remains the dominant gluconeogenic precursor, but amino acids such as alanine and glycerol from fat mobilization also contribute significantly. Emerging research using metabolomic profiling has shown that the efficiency of gluconeogenesis is highly variable among cows and may be genetically influenced, contributing to individual susceptibility to ketosis. This inter-animal variation highlights the importance of personalized nutritional strategies and the potential of genomic selection for improved metabolic resilience. The endocrine milieu of the transition cow is another crucial determinant of ketosis pathogenesis. Insulin plays a central role in regulating energy metabolism, yet dairy cows experience a state of peripheral insulin resistance during late gestation and early lactation. This physiological insulin resistance, which may have adaptive value in prioritizing glucose for the mammary gland, simultaneously promotes lipolysis and NEFA release from adipose tissue. Recent [9-12] studies have identified alterations in adipokine signaling, including reduced adiponectin and increased leptin resistance, which exacerbate the mobilization of fat reserves [3,16,20,40]. Moreover, the growth hormone–insulin-like growth factor-1 (GH–IGF-1) axis undergoes marked uncoupling, characterized by elevated circulating growth hormone but suppressed hepatic IGF-1 synthesis, further stimulating lipolysis while impairing anabolic processes. Thyroid hormones, cortisol, and other endocrine factors interact in this complex network, influencing not only energy partitioning but also immune competence and reproductive performance. Beyond the classical metabolic and hormonal frameworks, inflammation and oxidative stress are now recognized as integral

components of ketosis pathogenesis. Transition cows exhibit a systemic inflammatory response, even in the absence of overt infection, characterized by elevated acute-phase proteins and pro-inflammatory cytokines. Lipid mobilization and hepatic fatty infiltration potentiate oxidative stress, leading to mitochondrial dysfunction and further impairment of energy metabolism. Importantly, inflammatory mediators can suppress feed intake, aggravating NEB and perpetuating the cycle of metabolic stress [16]. Recent work has also highlighted the cross-talk between immune activation and hepatic metabolism, suggesting that immune-metabolic dysregulation may be a unifying concept underlying transition cow diseases. This paradigm shift underscores the need to view ketosis not merely as an isolated metabolic disorder but as a manifestation of broader immunometabolic dysfunction. The rumen microbiome has emerged as a novel factor influencing the development of ketosis. Shifts in microbial composition during the periparturient period can alter the production of volatile fatty acids, particularly propionate, which is the most important gluconeogenic substrate. Reduced abundance of propionate-producing bacteria has been associated with increased risk of subclinical ketosis, while microbial dysbiosis can also contribute to systemic inflammation through increased gut permeability and endotoxin translocation. These findings open the possibility that dietary or probiotic interventions targeting the rumen microbiota could play a role in mitigating ketosis risk, although this remains an emerging area of research requiring further validation [20]. From a systemic perspective, adipose tissue biology is increasingly recognized as central to transition cow physiology. Traditionally viewed only as an energy reserve, adipose tissue is now understood to be an active endocrine organ secreting adipokines, cytokines, and signaling molecules that influence hepatic metabolism, immune responses, and reproductive function. Recent histological and transcriptomic studies demonstrate that excessive and rapid lipolysis during the periparturient period induces adipose tissue inflammation, fibrosis, and impaired insulin signaling, establishing a feed-forward loop of metabolic dysfunction. This pathological remodeling of adipose tissue may help explain why cows with high body condition scores are particularly susceptible to ketosis and related disorders. Another emerging insight pertains to inter-individual variability in metabolic adaptation. Not all cows experiencing similar degrees of NEB develop ketosis, suggesting that genetic, epigenetic, and management-related factors confer resilience or vulnerability. Polymorphisms in genes involved in lipid metabolism, insulin signaling, and oxidative stress defense have been linked to differences in metabolic outcomes. Moreover, epigenetic modifications induced by nutritional and environmental stressors during the dry period may influence gene expression in key metabolic tissues. This growing body of knowledge supports the concept of “metabolic robustness” as a heritable trait and provides opportunities for selective breeding programs aimed at reducing the incidence of ketosis. The pathogenesis of ketosis cannot be fully understood without considering the interactions with other periparturient diseases. Cows with ketosis are at higher risk for displaced abomasum, retained placenta, metritis, and mastitis, and conversely, the presence of these disorders can exacerbate ketosis by reducing feed intake and increasing metabolic demands. These bidirectional relationships illustrate the complexity of the transition period, where metabolic, infectious, and reproductive disorders are interconnected components of a common pathophysiological network. Recognizing ketosis as part of a multifactorial disease complex has practical implications for herd health management, emphasizing integrated strategies rather than isolated interventions [3,16,20,40].

Taken together, the evolving picture of transition cow physiology and ketosis pathogenesis highlights the interplay of energy metabolism, endocrine regulation, immune function, and microbial ecology [21,32-34]. New insights from omics technologies, advanced imaging, and precision monitoring have shifted the narrative from a simplistic energy deficit model toward a multifactorial systems biology framework. This paradigm underscores that ketosis is not solely a consequence of inadequate nutrition or excessive milk yield, but rather the product of complex biological networks involving the liver, adipose tissue, mammary gland, immune system, and microbiome. Importantly, these discoveries provide avenues for innovation in prevention and management, ranging from targeted nutritional strategies and feed additives to genetic selection, microbiome modulation, and precision diagnostic tools [33]. Transition cow physiology represents a delicate balance between the metabolic demands of lactation and the homeostatic capacity of the

animal. When this balance is disrupted, the pathogenesis of ketosis emerges as a cascade involving NEB, excessive lipid mobilization, hepatic overload, impaired gluconeogenesis, endocrine dysregulation, inflammation, and microbial shifts. Recent advances have deepened our understanding of these processes, moving beyond descriptive observations to mechanistic insights at molecular, cellular, and systemic levels. These new perspectives not only refine our conceptual models of ketosis but also point toward novel biomarkers, intervention strategies, and genetic approaches that may improve metabolic health and productivity in dairy herds. Continued integration of molecular biology, precision technologies, and herd-level management will be essential in translating these insights into tangible improvements in the control of ketosis and the welfare of transition cows [21,32-34].

3. Abdominal adiposity, body condition scoring and metabolic risk

The relationship between body fat reserves, their distribution, and the risk of metabolic disorders such as ketosis has become a focal point of transition cow research in recent years [14-17]. Traditionally, dairy scientists and veterinarians have relied on body condition scoring (BCS) as a practical on-farm tool to assess energy reserves and predict health outcomes. BCS provides an inexpensive and rapid estimation of overall adiposity by visual and tactile assessment of anatomical landmarks, most often scored on a five-point scale. For decades, the prevailing consensus has been that overconditioned cows, particularly those calving with scores above 3.5 to 4.0, are at greater risk of excessive lipid mobilization, negative energy balance, and subsequent ketosis. While this principle remains valid, new insights emphasize that not only the total amount of fat but also its distribution—especially abdominal and visceral adiposity—plays a pivotal role in determining metabolic risk. Adipose tissue is not simply an inert energy depot but an active endocrine and immune organ, secreting adipokines, cytokines, and metabolites that influence systemic metabolism. Accumulation of abdominal fat, particularly intra-abdominal and omental depots, has been linked to adverse metabolic outcomes in both humans and cattle [1,15]. Unlike subcutaneous fat, visceral adipose tissue exhibits greater lipolytic activity, higher sensitivity to catecholamines, and reduced responsiveness to insulin. This means that cows with disproportionate abdominal adiposity may mobilize NEFA more aggressively in early lactation, overwhelming hepatic oxidative capacity and predisposing to ketosis and fatty liver. Recent studies employing ultrasonography, computed tomography, and carcass dissections have provided quantitative evidence that cows with higher visceral fat content, independent of overall BCS, show elevated postpartum NEFA and beta-hydroxybutyrate concentrations. These findings challenge the traditional reliance on external body condition alone as a predictor of metabolic health. One of the main limitations of classical BCS is its subjective nature and focus on subcutaneous reserves. Two cows with identical scores can differ markedly in the amount of internal adiposity, leading to divergent metabolic risks despite similar external appearance. This heterogeneity partly explains why not all overconditioned cows develop ketosis and why some apparently moderate-conditioned cows still show severe postpartum metabolic derangements.

New imaging [14-17] approaches have therefore been explored to better capture abdominal fat reserves [14,38-40]. Ultrasonographic assessment of perirenal and omental fat thickness, for example, has shown good correlation with metabolic markers. Portable ultrasound devices make this approach increasingly feasible in field conditions, though it remains more time-consuming than traditional scoring. Infrared thermography and bioelectrical impedance are also under investigation as non-invasive techniques for estimating body fat distribution. Integration of such objective tools with traditional BCS could substantially improve risk stratification. Beyond structural quantification, the metabolic activity of adipose tissue has come under intense scrutiny. In overconditioned cows, adipocytes enlarge (hypertrophy), leading to local hypoxia, recruitment of macrophages, and secretion of pro-inflammatory mediators. This state of adipose tissue inflammation contributes to systemic low-grade inflammation observed in transition cows. Elevated circulating cytokines such as TNF- α and IL-6 can exacerbate insulin resistance and suppress feed

intake, both of which intensify negative energy balance. Furthermore, inflamed adipose tissue exhibits impaired insulin signaling pathways, reducing the capacity to suppress lipolysis even in the presence of insulin. As a result, visceral fat depots in particular become metabolically unstable during the periparturient period, releasing large quantities of NEFA into circulation and fueling hepatic ketogenesis. These mechanistic insights position abdominal adiposity not just as a passive risk factor but as an active driver of metabolic dysregulation. Another important consideration is the dynamic change in BCS rather than static values at calving. Excessive loss of condition in early lactation, often defined as more than 0.5 to 0.75 units within the first month postpartum, has consistently been associated with higher incidence of ketosis, displaced abomasum, and reduced reproductive performance. Rapid mobilization of adipose tissue indicates both insufficient dietary energy intake and heightened catabolic drive. The interplay between calving BCS, abdominal adiposity, and postpartum BCS loss is complex. Overconditioned cows tend to lose more condition after calving, mobilize more visceral fat, and are therefore doubly disadvantaged. Conversely, cows calving too thin (<2.5 BCS) may lack adequate reserves to support lactation, leading to poor milk yield and compromised fertility. These observations reinforce the importance of achieving an optimal, moderate body condition at calving, typically around 3.0 to 3.25, while minimizing both overconditioning and excessive postpartum mobilization. Genetic and epigenetic factors also modulate the relationship between adiposity and metabolic risk. Some cows appear to tolerate higher BCS without adverse outcomes, while others exhibit disproportionate susceptibility to ketosis despite moderate scores. Polymorphisms in genes regulating lipolysis, fatty acid oxidation, and insulin signaling may underlie these differences. For example, variations in perilipin and hormone-sensitive lipase genes have been associated with altered lipolytic responses. Moreover, nutritional programming during the dry period can epigenetically influence adipose tissue biology, potentially predisposing offspring and subsequent lactations to altered fat metabolism. These emerging findings suggest that a one-size-fits-all approach to BCS targets may not be optimal and that herd- or cow-specific strategies may be required. From a management perspective, abdominal adiposity and BCS remain practical levers for controlling metabolic disease risk, but their application requires nuance. Overfeeding during the dry period, especially of energy-dense diets, remains the primary cause of overconditioning. Controlled-energy or high-fiber dry cow diets have been shown to reduce the risk of excessive fat accumulation and improve postpartum energy balance. However, new work emphasizes that the timing of nutritional interventions matters as much as diet composition. Cows that gain excessive condition in late lactation are particularly vulnerable, underscoring the importance of monitoring throughout the production cycle rather than focusing exclusively on the dry period. Exercise, housing comfort, and grouping strategies can also influence feed intake and condition dynamics, further highlighting the multifactorial nature of adiposity control. Economic and welfare implications of abdominal adiposity are increasingly recognized. Overconditioned cows not only suffer higher incidence of ketosis but also reduced reproductive efficiency, longer days open, and greater culling risk. From an economic standpoint, the costs of lost milk yield, treatment, and replacement outweigh the expense of preventive management.

From a welfare perspective [14,38-40], cows undergoing excessive lipid mobilization experience physiological stress that predisposes to concurrent diseases such as metritis and mastitis [14,45,48,49]. Addressing adiposity therefore aligns both with herd profitability and animal welfare objectives. Integration of novel technologies may refine how abdominal adiposity and BCS are used in practice. Automated camera systems are being developed to provide continuous digital body condition scoring with high accuracy, minimizing subjective bias and enabling large-scale herd monitoring. When combined with milk yield data, feed intake records, and sensor-based activity measurements, such systems could provide real-time risk assessment models for ketosis. Emerging biomarkers, such as circulating adipokines or lipidomic signatures, may further enhance predictive capacity when integrated with structural measures of adiposity. The ultimate goal is to move toward precision management, where individual cows are monitored and managed according to their specific risk profiles rather than herd averages. Abdominal adiposity and body condition scoring are central determinants of metabolic risk in transition dairy cows, but their interpretation has evolved

substantially in light of new evidence. It is no longer sufficient to consider only external condition; the distribution, metabolic activity, and inflammatory status of adipose depots are equally critical. Visceral fat plays a disproportionately harmful role due to its lipolytic sensitivity and endocrine activity, making it a major driver of ketosis pathogenesis. Traditional BCS remains a valuable tool, but its limitations necessitate supplementation with more objective and nuanced measures, such as ultrasonography, digital imaging, and biochemical markers. Effective management requires attention to both the prevention of overconditioning and the avoidance of excessive postpartum mobilization, with nutritional, genetic, and technological strategies all contributing to improved outcomes [14,45,48,49]. As the dairy industry moves toward data-driven precision management, the integration of adiposity assessment into predictive models of metabolic disease will be crucial. Ultimately, the refinement of our understanding of body condition and fat distribution not only enhances metabolic health and productivity but also promotes animal welfare and sustainability in modern dairy production systems (Table 1, Figure 1).

Table 1. Risk factors associated with the development of ketosis in transition dairy cows.

Risk factor	Effect on ketosis risk	Relative risk
BCS < 2.5	Low energy reserves	Moderate
BCS 3.0–3.25	Optimal condition	Low
BCS > 3.75	Excessive lipolysis postpartum	High
Excessive NEFA mobilization	Increased ketogenesis	High
Reduced dry matter intake	Negative energy balance	High
Multiparity	Higher metabolic load	Moderate
Heat stress	Reduced feed intake	Moderate
Rumen dysbiosis	Altered VFA production	Moderate–High

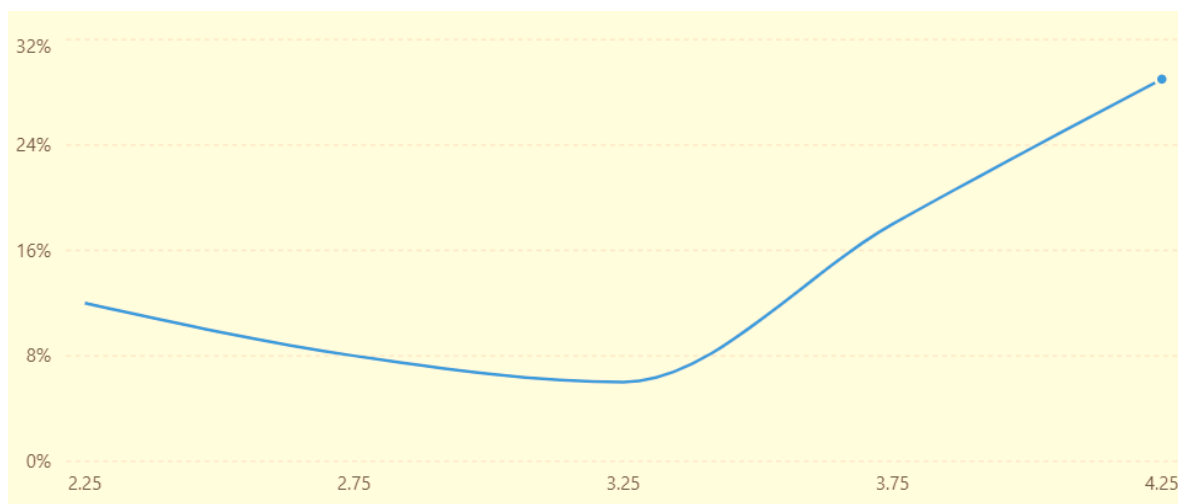


Figure 1. Relationship between body condition score (BCS) at calving and ketosis incidence in dairy cows.

The lowest incidence is observed in cows calving with a moderate BCS (3.0–3.25), whereas both underconditioned and overconditioned cows exhibit increased metabolic risk.

4. Multi-omics (metabolomics and lipidomics) signatures of ketosis

The study of ketosis in dairy cattle has traditionally relied on measurements of blood β -hydroxybutyrate (BHB), non-esterified fatty acids (NEFA), and glucose as the primary biochemical markers [21-24]. While these indicators remain central to the diagnosis and monitoring of the disease, they capture only a small fraction of the complex metabolic derangements underlying

ketosis. In recent years, the development of high-throughput analytical platforms, including nuclear magnetic resonance (NMR), liquid chromatography–mass spectrometry (LC-MS), gas chromatography–mass spectrometry (GC-MS), and targeted lipidomic assays, has enabled unprecedented insights into the metabolic landscape of transition cows. Metabolomics and lipidomics, as key branches of the multi-omics paradigm, provide comprehensive snapshots of small molecules and lipid species, respectively, allowing researchers to unravel novel biomarkers, mechanistic pathways, and therapeutic targets associated with ketosis. One of the central findings from metabolomic investigations is that the metabolic signature of ketosis extends well beyond the elevation of ketone bodies. Cows in early lactation that develop subclinical or clinical ketosis exhibit profound perturbations in amino acid metabolism, tricarboxylic acid (TCA) cycle intermediates, bile acids, and gut-derived metabolites. Depletion of glucogenic amino acids such as alanine, glycine, and serine is frequently reported, reflecting their increased utilization as precursors for hepatic gluconeogenesis during periods of negative energy balance. At the same time, metabolites associated with impaired TCA cycle activity, including citrate and succinate, are reduced, suggesting a bottleneck in oxidative metabolism and a shift toward incomplete oxidation of fatty acids. This impaired mitochondrial function is thought to exacerbate the accumulation of acetyl-CoA, subsequently fueling ketogenesis [21-24].

Metabolomic profiling has also identified novel markers that are strongly correlated with the risk of ketosis and may precede the appearance of clinical symptoms [21-24]. Compounds such as acylcarnitines, trimethylamine N-oxide (TMAO), and specific bile acids have been consistently associated with lipid mobilization and hepatic stress. Elevated levels of medium- and long-chain acylcarnitines are particularly notable, as they reflect mitochondrial overload and incomplete fatty acid oxidation in the liver [23]. These findings have positioned acylcarnitines as promising early-warning biomarkers that integrate both adipose tissue lipolysis and hepatic metabolic capacity. In parallel, lipidomics has significantly expanded the understanding of how lipid species dynamics contribute to the pathogenesis of ketosis. Beyond total NEFA concentration, lipidomic studies reveal that the composition of circulating fatty acids and complex lipids changes markedly during the transition period. Polyunsaturated fatty acids (PUFAs), phosphatidylcholines, and sphingolipids are altered in cows developing ketosis, suggesting that membrane remodeling, inflammatory signaling, and lipoprotein metabolism are intricately involved in disease progression. Importantly, specific lipid signatures, such as reduced levels of lysophosphatidylcholines and altered ratios of ceramides to sphingomyelins, have been linked to both metabolic inflammation and impaired insulin sensitivity in transition cows. These lipidomic alterations mirror mechanisms described in human metabolic disorders, underscoring the cross-species relevance of lipid dysregulation in energy metabolism. Another key insight from lipidomics is the heterogeneity of NEFA mobilization. Not all fatty acids contribute equally to ketosis risk: saturated fatty acids such as palmitic acid appear to impose greater metabolic strain on the liver than unsaturated fatty acids. Furthermore, lipidomic fingerprints have revealed that cows with excessive adiposity mobilize distinct lipid profiles compared to leaner cows, emphasizing that body condition not only affects the magnitude but also the quality of lipid mobilization. Such differences in lipid composition may partially explain why some cows tolerate high NEFA concentrations without developing clinical ketosis, whereas others succumb rapidly despite lower NEFA burdens [23]. Integrative multi-omics approaches that combine metabolomic and lipidomic data with transcriptomic or proteomic layers are providing deeper mechanistic insights. For example, studies linking metabolite and lipid signatures with hepatic gene expression demonstrate that perturbations in the peroxisome proliferator-activated receptor (PPAR) signaling pathway are central to ketosis pathogenesis. Downregulation of genes involved in fatty acid β -oxidation coincides with accumulation of incomplete oxidation products, while altered expression of lipid transport proteins parallels changes in plasma lipoprotein profiles detected by lipidomics. Such cross-level correlations strengthen the causal inference that mitochondrial dysfunction and impaired lipid trafficking are not merely consequences but active drivers of ketosis development. From a practical standpoint, the application of multi-omics to ketosis research is paving the way toward precision livestock health management [24]. Metabolomic and lipidomic signatures are increasingly being tested as predictive tools, aiming

to identify cows at risk of ketosis before calving or in the very early postpartum period. Predictive panels incorporating combinations of acylcarnitines, specific amino acids, and phospholipid species have demonstrated higher sensitivity and specificity than traditional markers such as BHB and NEFA alone. This raises the possibility of developing metabolomic-based screening assays that could be integrated into routine herd health programs. Furthermore, multi-omics data are enhancing the understanding of how nutrition, genetics, and management interact in determining ketosis susceptibility. For instance, metabolomic profiling has shown that cows supplemented with rumen-protected choline or specific fatty acid blends exhibit distinct plasma metabolite patterns that correlate with improved hepatic lipid export and reduced ketosis incidence. Similarly, genetic lines selected for feed efficiency display unique lipidomic fingerprints that may underlie their resilience to metabolic stress. Such findings highlight the potential of omics-guided nutritional and breeding strategies to mitigate the burden of ketosis at the herd level. The emerging role of the gut microbiome in shaping the metabolomic landscape of transition cows adds another layer of complexity.

Gut-derived metabolites such as short-chain fatty acids [21-24] (SCFAs), indole derivatives, and secondary bile acids are increasingly recognized as modulators of hepatic metabolism and systemic inflammation [21,23-25]. Multi-omics studies integrating fecal microbiota composition with circulating metabolome data suggest that microbial dysbiosis during the transition period may contribute to altered energy partitioning and increased ketosis risk. These insights pave the way for microbiome-targeted interventions, including probiotics and dietary fiber supplementation, as adjuncts to conventional ketosis prevention strategies. Despite these advances, several challenges remain before multi-omics approaches can be fully translated into practical applications. The high dimensionality of metabolomic and lipidomic datasets requires sophisticated statistical and machine-learning models for interpretation, and results may vary depending on analytical platform, sample preparation, and data processing methods. Standardization of protocols and the establishment of large-scale reference databases are critical to ensure reproducibility and cross-study comparability. Moreover, while many metabolomic biomarkers have been identified, their causal roles in ketosis remain to be validated through mechanistic experiments. Future work will need to bridge the gap between correlation and causation, possibly through functional studies using isotope tracing, targeted knockout models, or intervention trials. The integration of metabolomics and lipidomics has revolutionized the understanding of ketosis in dairy cattle by revealing complex and dynamic signatures that extend beyond traditional biomarkers. These approaches highlight the multifactorial nature of ketosis, encompassing impaired mitochondrial function, altered lipid mobilization, amino acid depletion, and gut-liver cross-talk. The ongoing shift toward multi-omics frameworks promises not only to refine diagnostic accuracy but also to open novel avenues for prevention and individualized management of ketosis. As analytical technologies become more accessible and data integration methods mature, it is expected that metabolomic and lipidomic signatures will move from experimental research toward routine application in precision dairy health, ultimately contributing to improved animal welfare, productivity, and farm profitability [21,23-25].

5. Rumen microbiome and metabolite–host interactions in postpartum ketosis

The rumen microbiome has emerged as a central determinant of dairy cow health and productivity, particularly during the vulnerable transition period [28-31]. While traditionally the pathogenesis of ketosis has been attributed mainly to excessive lipid mobilization and impaired hepatic metabolism, recent research has uncovered that microbial populations and their metabolic outputs in the rumen strongly influence the host's energy balance, immune function, and susceptibility to metabolic disease. The concept of ketosis as a disorder involving not only the liver and adipose tissue but also microbial–host interactions reflects a major paradigm shift in ruminant metabolic research [30]. Advances in next-generation sequencing, metagenomics, and metabolomics have provided detailed characterizations of microbial communities and their metabolites, enabling

the integration of rumen microbiota composition with systemic metabolic signatures. This new perspective has expanded the understanding of postpartum ketosis beyond traditional host-centric models and has identified the microbiome as both a contributor to disease risk and a potential therapeutic target. One of the most consistent observations is that the rumen microbiome undergoes substantial remodeling around parturition, a period marked by dramatic shifts in diet, endocrine status, and nutrient demands. The transition from a dry cow diet rich in forage to a lactation diet with higher starch content alters microbial diversity and metabolic outputs. Cows that subsequently develop ketosis frequently exhibit reduced microbial richness and an altered Firmicutes-to-Bacteroidetes ratio [31]. Specifically, decreases in fiber-degrading genera such as *Ruminococcus* and increases in starch-utilizing bacteria like *Prevotella* have been associated with excessive production of propionate and lactate, which may overwhelm metabolic buffering systems. Conversely, reduced abundance of butyrate-producing microbes limits the generation of this key energy source for rumen epithelium, thereby compromising epithelial integrity and nutrient absorption.

These changes [28-31] collectively create an environment that predisposes cows to suboptimal fermentation patterns and altered delivery of volatile fatty acids (VFAs) to the host [28-31]. VFAs are the primary metabolic link between the rumen microbiota and the host, providing up to 70% of the cow's energy requirements. Altered VFA profiles in cows at risk of ketosis include decreased acetate-to-propionate ratios and increased levels of branched-chain VFAs. Such changes affect systemic glucose supply because propionate is the major gluconeogenic precursor. An imbalance favoring excessive propionate production can paradoxically impair energy homeostasis if hepatic gluconeogenesis is compromised by concurrent lipid overload. Furthermore, shifts in VFA absorption dynamics, influenced by the integrity of rumen epithelium and the expression of transporter proteins, may exacerbate mismatches between nutrient supply and demand. This interplay highlights that the microbiome not only dictates what metabolites are produced but also indirectly modulates how efficiently the host can utilize them. Beyond VFAs, other microbial metabolites have gained attention as potential mediators of ketosis risk. Microbial-derived amino acid catabolites, indoles, and phenolic compounds influence systemic inflammatory tone and hepatic detoxification pathways. Elevated levels of p-cresol, indole derivatives, and ammonia have been reported in cows prone to metabolic stress, suggesting that microbial dysbiosis increases the burden of potentially harmful metabolites that compete with hepatic energy metabolism. Secondary bile acids, generated by microbial transformation of host bile salts, also play a role by activating signaling pathways such as farnesoid X receptor (FXR) and Takeda G protein-coupled receptor 5 (TGR5), which regulate glucose and lipid metabolism. Dysregulated bile acid signaling, driven by microbial imbalance, may therefore contribute to impaired hepatic adaptation and promote ketogenesis. The rumen epithelium itself serves as an active interface where microbial metabolites interact with host physiology. Short-chain fatty acids, particularly butyrate, modulate epithelial gene expression through histone deacetylase inhibition, influencing barrier function, immune surveillance, and transport capacity. In cows that later develop ketosis, reduced availability of butyrate is associated with downregulation of epithelial tight junction proteins and increased permeability, facilitating the translocation of bacterial components such as lipopolysaccharide (LPS). Elevated systemic LPS, a hallmark of subacute ruminal acidosis and dysbiosis, contributes to inflammatory activation, impaired insulin signaling, and diversion of hepatic resources away from gluconeogenesis toward detoxification and acute-phase responses. This microbial-host crosstalk suggests that immune-metabolic stress originating from the rumen may act synergistically with negative energy balance to accelerate the onset of ketosis. Metabolomic analyses linking rumen fluid composition to circulating blood profiles provide direct evidence for the systemic impact of microbial metabolites. Cows with ketosis exhibit elevated plasma concentrations of microbial-derived metabolites such as trimethylamine N-oxide (TMAO), which reflects altered microbial choline metabolism and is associated with hepatic lipid accumulation. Similarly, shifts in branched-chain amino acid catabolism within the rumen are mirrored by altered circulating levels of valine, leucine, and isoleucine, metabolites that have been implicated in insulin resistance in both cattle and humans. Such parallels underscore the relevance of microbial activity in modulating host

metabolic resilience or vulnerability during the transition period. The recognition of the rumen microbiome's role in ketosis pathogenesis has spurred exploration of microbiome-targeted interventions. Nutritional strategies such as the use of prebiotics, probiotics, and direct-fed microbials are being evaluated for their capacity to stabilize microbial populations, promote butyrate production, and mitigate dysbiosis. For instance, supplementation with specific strains of *Lactobacillus*, *Propionibacterium*, or *Megasphaera elsdenii* has been shown to modulate VFA profiles, reduce ruminal lactate accumulation, and improve systemic energy balance. Similarly, dietary fiber fractions that selectively enhance the growth of fibrolytic microbes may restore microbial diversity and reinforce epithelial barrier integrity. Beyond nutritional manipulation, fecal microbiota transplantation has emerged as a novel experimental approach, suggesting that deliberate reshaping of microbial communities may confer metabolic benefits in high-risk cows [28-31].

Another promising avenue lies in the integration of microbiome data with host genomic and transcriptomic profiles, which may reveal host-microbe interactions that determine individual susceptibility to ketosis [28-30]. Variations in host genes encoding VFA transporters, immune receptors, and hepatic enzymes interact with microbial outputs to shape metabolic outcomes. Precision livestock farming technologies are beginning to incorporate microbial biomarkers into predictive models, allowing earlier identification of cows likely to develop ketosis based on their rumen microbial signatures. Such predictive capacity opens the door to personalized nutritional and management interventions tailored to individual microbial and metabolic profiles. Despite significant progress, several challenges remain in translating microbiome research into practical ketosis control strategies. The rumen microbiome is highly dynamic, influenced by diet, environment, and host genetics, making it difficult to define stable biomarkers. Furthermore, causality is often difficult to establish, as microbial changes may be both a cause and a consequence of metabolic imbalance. Standardization of sampling protocols, sequencing methods, and bioinformatic pipelines is urgently needed to ensure reproducibility and comparability across studies. Longitudinal studies tracking microbial shifts before, during, and after ketosis episodes will be crucial to disentangle temporal relationships and identify robust microbial predictors of disease. The rumen microbiome and its metabolites are now recognized as central players in the pathogenesis of postpartum ketosis, acting through diverse mechanisms that include modulation of VFA supply, alteration of bile acid and amino acid metabolism, disruption of epithelial barrier integrity, and systemic immune activation. The host, in turn, shapes the microbiome through diet, immunity, and endocrine signals, creating a bidirectional interaction that can either support adaptation to lactation or precipitate metabolic collapse. By embracing this systems-level perspective, future strategies for ketosis prevention and management will likely move beyond liver-focused interventions to include microbiome-directed approaches, advancing both cow health and farm profitability [28-30].

6. Precision monitoring and sensor integration for early detection

The early detection of ketosis in dairy cattle is one of the most critical aspects of disease prevention and herd health management [46-49]. Historically, diagnosis has relied on point measurements of blood β -hydroxybutyrate (BHB), milk ketones, or urine dipsticks, all of which are episodic, labor-intensive, and limited by the timing of sampling. However, the emergence of precision livestock farming (PLF) technologies and sensor-based monitoring systems has revolutionized the capacity to detect metabolic disturbances in real time. The integration of wearable, in-line, and environmental sensors, combined with advanced data analytics and machine learning algorithms, is transforming ketosis detection from a reactive to a predictive discipline, where at-risk cows can be identified before clinical signs develop. Modern precision monitoring systems capture a wide spectrum of physiological, behavioral, and production-related parameters that serve as proxies for metabolic health. Rumen boluses and collars equipped with accelerometers and gyroscopes provide continuous data on rumination, eating behavior, and physical activity.

Subtle declines in rumination time, reduced feeding bouts, or increased lying time often precede biochemical evidence of ketosis, reflecting the cow's early adaptive response to energy imbalance. Similarly, automated feed intake monitoring systems record deviations in dry matter intake at the individual cow level, which are strongly correlated with postpartum negative energy balance and subsequent risk of ketosis. Such systems surpass the resolution of traditional pen-level monitoring, enabling the detection of within-herd variability and identification of individuals most vulnerable to metabolic failure. In parallel, milk sensors embedded in robotic milking systems or parlor-based platforms have opened new opportunities for metabolic monitoring. Infrared spectroscopy of milk provides information not only on fat, protein, and lactose content but also on subtle changes in fatty acid profiles that indicate excessive lipid mobilization. Ratios such as fat-to-protein or fat-to-lactose, calculated from routine milk testing, have been validated as indirect markers of ketosis risk [46-49].

More recently, sensors capable of measuring milk BHB directly at the point of milking have been commercialized, allowing continuous tracking of ketone dynamics at the udder level [49,50,78,79]. When coupled with other milk traits, these sensors provide high sensitivity and specificity for subclinical ketosis, a condition that often remains undetected by farmers but carries substantial production and fertility penalties. Wearable sensors measuring physiological parameters further enrich the monitoring landscape. Reticulo-rumen boluses can measure core body temperature and pH, indirectly reflecting fermentation efficiency and the risk of ruminal acidosis, which often co-occurs with energy metabolism disorders. Ear tag sensors record activity and position data, which when combined with feeding information, yield composite indicators of metabolic stress. More advanced biosensors are now capable of monitoring metabolites such as glucose, NEFA, or BHB in interstitial fluid through minimally invasive techniques, resembling continuous glucose monitoring in human medicine. Although still in experimental stages, these biosensors promise to provide direct biochemical markers in real time, potentially replacing traditional sampling altogether. The integration of multiple sensor data streams requires sophisticated data processing and interpretation frameworks. Machine learning and artificial intelligence (AI) approaches are increasingly applied to sensor-derived datasets to identify patterns predictive of ketosis. Algorithms trained on large-scale datasets incorporating behavior, milk yield, milk composition, and activity can achieve predictive accuracies exceeding those of single-parameter systems. For example, models that integrate feeding behavior, rumination, and milk yield trajectories can detect cows at risk of ketosis up to one week before clinical diagnosis, allowing for timely intervention. Importantly, these predictive models continue to improve as they are trained on larger, farm-specific datasets, adapting to differences in management, nutrition, and genetics. An additional dimension of precision monitoring lies in the integration of environmental and management-related data. Sensors that track barn climate, temperature-humidity index (THI), and lying surface comfort contribute valuable context to metabolic monitoring. Heat stress, for example, exacerbates negative energy balance and alters microbial fermentation, thereby increasing ketosis risk. By incorporating environmental data, sensor platforms can distinguish between behavioral changes caused by heat stress versus those attributable to metabolic disorders, reducing false positives and improving the specificity of alerts. Beyond individual sensors, the real innovation lies in multi-sensor integration platforms. Cloud-based herd management systems now combine information from milking robots, rumen boluses, accelerometers, and feeding stations into unified dashboards accessible to farmers and veterinarians. These platforms provide not only individual cow alerts but also herd-level insights, such as clustering of ketosis cases, correlations with nutritional management, or patterns linked to calving season. By contextualizing ketosis risk within herd dynamics, such systems support proactive management decisions, including ration adjustments, regrouping strategies, or targeted supplementation programs [49,50]. Economic considerations are central to the adoption of precision monitoring systems. While sensor technologies require substantial investment, economic modeling consistently demonstrates favorable cost-benefit ratios due to reductions in milk yield losses, improved fertility outcomes, and decreased veterinary costs associated with ketosis and its sequelae. Early detection minimizes the progression from subclinical to clinical disease, reduces the need for therapeutic interventions, and improves overall longevity and productivity. Furthermore, as the cost of sensors continues to

decline and their robustness improves, widespread adoption across dairy systems of varying scales is expected [78,79]. The integration of precision monitoring into ketosis management also aligns with broader sustainability and animal welfare goals. Continuous monitoring reduces the need for invasive blood sampling and minimizes handling stress, improving animal welfare. Data-driven detection allows interventions to be tailored and targeted, reducing unnecessary treatments and promoting judicious use of veterinary resources. Moreover, precision monitoring enhances transparency in animal health management, which is increasingly demanded by consumers and regulatory frameworks emphasizing animal welfare and sustainability [49,50,78,79].

Despite significant progress, several challenges remain in fully realizing the potential of sensor-based ketosis monitoring [46-49]. One major limitation is the standardization and interoperability of devices from different manufacturers. Data silos created by proprietary systems hinder seamless integration, limiting the ability to generate holistic insights. Furthermore, false positives remain a concern, particularly when single-parameter thresholds are used without accounting for the complex physiology of transition cows. Machine learning models mitigate this issue but require ongoing validation and calibration under diverse farm conditions. Data privacy and ownership also pose challenges, as farmers and technology providers negotiate the control and sharing of large-scale livestock datasets. Looking forward, the integration of sensor-based monitoring with other omics technologies represents a promising frontier. Linking metabolomic and lipidomic signatures of ketosis with real-time behavioral and physiological data could enable mechanistic, rather than correlative, predictions of disease risk. Such integration would allow detection systems not only to identify which cows are at risk but also to provide insights into the underlying metabolic pathways involved, thereby informing more precise interventions. Personalized health management strategies could emerge, where cows are managed individually based on their unique metabolic, behavioral, and microbial profiles. Precision monitoring and sensor integration are reshaping the approach to ketosis detection in dairy cows, shifting from episodic, reactive diagnosis to continuous, predictive surveillance [46-49]. Wearable, in-line, and environmental sensors generate rich datasets that, when combined through advanced analytics, enable earlier detection of metabolic imbalance, improved specificity of alerts, and proactive herd management. While technical and logistical challenges remain, the ongoing convergence of precision farming, data science, and veterinary medicine promises to make early ketosis detection not only more accurate but also more economically and ethically sustainable. The widespread adoption of these technologies is likely to represent one of the most transformative advances in the prevention and management of metabolic diseases in dairy cattle in the coming decade (Table 2).

Table 2. Emerging technologies for early detection of ketosis in dairy cattle.

Technology	Measured parameter	Detection potential
Milk BHB sensor	Ketone bodies	Very high
Activity collar	Activity and rumination	High
Rumen bolus	Temperature and pH	Moderate
Infrared milk spectroscopy	Milk composition	High
AI prediction models	Multiple variables	Very high
Biosensors	Glucose, NEFA, BHB	Experimental

7. Interventions: nutritional, pharmaceutical and microbiome-targeted strategies

The prevention and management of ketosis in dairy cattle relies on interventions that target multiple physiological systems, reflecting the multifactorial nature of the disorder [37,42-44]. While nutritional approaches remain the cornerstone of ketosis control, advances in pharmaceutical treatments and emerging microbiome-targeted strategies are expanding the range of available tools. The transition from traditional reactive therapy toward preventive and individualized intervention frameworks is supported by recent insights into the physiology, pathogenesis, and microbial–host

interactions that underlie the disease. A comprehensive overview of these intervention strategies illustrates both the progress made and the challenges that remain in effectively mitigating ketosis in modern dairy systems. Nutritional management is the first line of defense against ketosis, given that the disease originates from an imbalance between energy supply and demand in early lactation. Strategies to optimize dry matter intake before and after calving are critical, as sufficient intake buffers cows against negative energy balance. Prepartum diets are formulated to avoid excessive energy density, preventing overconditioning and limiting adipose tissue mobilization postpartum. Controlled-energy diets for dry cows, often based on high-fiber, low-starch rations, have consistently reduced the incidence of ketosis by moderating body condition score gain during the dry period. Postpartum, maximizing feed intake through palatable, frequent feeding and careful ration formulation ensures the provision of adequate glucose precursors [37]. Carbohydrate sources that favor propionate production in the rumen, such as starch from finely ground corn or barley, directly support hepatic gluconeogenesis and help counteract ketone accumulation. Nutrient supplementation represents another critical nutritional intervention [42-44]. Glucogenic precursors such as propylene glycol remain one of the most widely used preventive tools administered orally in the days immediately after calving to stimulate hepatic gluconeogenesis and lower circulating BHB [42-45]. Glycerol has a similar role, though its efficacy is somewhat less consistent. Niacin supplementation has been explored as a lipolysis inhibitor, reducing NEFA mobilization, though results are variable depending on dose and diet composition. Choline, particularly in rumen-protected form, enhances very-low-density lipoprotein (VLDL) export from the liver, mitigating fatty liver and indirectly reducing ketosis risk. Methionine, another methyl donor, is increasingly recognized for its role in hepatic lipid metabolism, with studies demonstrating reduced liver triacylglycerol accumulation when supplemented prepartum. Trace minerals such as chromium and selenium, and vitamins such as vitamin E, contribute to improved insulin sensitivity and antioxidant defense, thereby supporting metabolic resilience during the transition period. Dietary fat manipulation has also been investigated as a means of improving energy supply without excessive fermentation load. Rumen-protected fats provide a concentrated source of energy, but the type of fatty acid supplied matters. Saturated fats such as palmitic acid may exacerbate hepatic lipid accumulation, whereas unsaturated fatty acids, particularly omega-3 fatty acids, appear to support hepatic lipid oxidation and reduce inflammation. Balancing carbohydrate and fat supply is therefore critical to avoid shifting the metabolic burden from glucose deficiency to lipid overload. Pharmaceutical interventions, although not a substitute for sound nutrition, play an important role in both prevention and treatment. Propylene glycol is often considered both a nutritional and pharmaceutical tool given its mode of administration and rapid metabolic effects. Intravenous glucose remains the primary emergency treatment for clinical ketosis, quickly reducing ketone body concentrations, although its effects are short-lived without concurrent oral supplementation. Dexamethasone, a glucocorticoid, has been employed to stimulate appetite and gluconeogenesis, though concerns about immunosuppression and reproduction limit its routine use. Insulin therapy, particularly long-acting formulations combined with glucose infusion, has shown efficacy in restoring energy balance, but its cost and practical limitations restrict widespread use. Monensin, delivered through controlled-release capsules, alters rumen fermentation to increase propionate production, thereby reducing ketosis incidence at the herd level. While effective, regulatory restrictions and consumer preferences in some regions limit its universal application. Beyond conventional pharmaceutical options, interest is growing in hepatoprotective and metabolic-modulating agents. Compounds that enhance mitochondrial function, such as L-carnitine, have been explored to improve fatty acid oxidation, though results remain mixed. Emerging pharmaceutical approaches include the use of peroxisome proliferator-activated receptor (PPAR) agonists, modeled after human diabetes therapies, to improve lipid handling and reduce ketogenesis. While experimental at this stage, such targeted metabolic modulators may offer future avenues for intervention as regulatory and safety evaluations progress. A rapidly evolving frontier in ketosis management involves microbiome-targeted interventions. Recognition of the rumen microbiome as a key determinant of host energy metabolism has driven efforts to manipulate microbial communities to support metabolic stability. Probiotics containing lactic acid-utilizing or

butyrate-producing strains, such as *Megasphaera elsdenii* or certain *Lactobacillus* and *Bifidobacterium* species, have been tested with promising effects on rumen fermentation profiles. Direct-fed microbials can enhance fiber digestion, stabilize rumen pH, and promote VFA profiles that favor gluconeogenesis and reduce lipid mobilization. Prebiotics, including specific oligosaccharides and non-digestible fibers, selectively enrich beneficial microbial populations, supporting microbial diversity and resilience during the transition period. Yeast cultures, particularly *Saccharomyces cerevisiae*, are another widely used microbiome-targeted strategy. Their effects include increased rumen pH stability, enhanced fiber degradation, and stimulation of lactate-utilizing bacteria, all of which indirectly reduce the risk of subacute ruminal acidosis and associated energy imbalances. By promoting a more favorable microbial environment, yeast supplementation can support greater dry matter intake and milk production while reducing ketosis incidence. Microbiome modulation also extends to the use of plant-derived bioactive compounds with antimicrobial and anti-inflammatory properties.

Essential oils, tannins, and saponins can shift [42-45], microbial populations, suppress pathogens, and reduce systemic inflammatory load, which otherwise impairs hepatic metabolism during early lactation [55-58]. The integration of these natural products into rations reflects the growing interest in alternatives to antibiotics and ionophores in livestock health management. Fecal microbiota transplantation (FMT), though experimental, represents a novel microbiome-targeted intervention. Early trials suggest that transferring microbial communities from metabolically healthy donors to at-risk cows may restore microbial balance and improve energy partitioning. While practical challenges and biosecurity concerns must be addressed, the potential of FMT underscores the central role of the microbiome in shaping host metabolic outcomes. An important consideration across all intervention types is the need for integration and timing.

Nutritional, pharmaceutical, and microbiome-targeted strategies are most effective when combined within a holistic management framework rather than applied in isolation. For example, propylene glycol drenching is more successful when accompanied by controlled prepartum diets and microbiome stabilization [55].

Similarly, choline supplementation demonstrates its greatest impact in cows with high body condition scores and concurrent microbial dysbiosis. The shift toward precision livestock farming enables interventions to be applied in a targeted, individualized manner, informed by sensor-based monitoring and omics-derived biomarkers. By aligning the type of intervention with the cow's specific metabolic and microbial profile, the efficacy of prevention strategies is maximized while unnecessary treatments are minimized. While the range of available interventions is expanding, barriers to adoption remain.

Economic considerations are critical, as high-cost supplements or pharmaceutical treatments may not be feasible for all farming systems. Regulatory approval processes and consumer concerns about pharmaceutical residues also shape the availability of certain tools across regions [56]. Moreover, variability in response to interventions reflects the heterogeneity of cows, farms, and environments, highlighting the need for ongoing research to refine recommendations and develop predictive models that identify which cows will benefit most from specific strategies [57].

Interventions for ketosis encompass a spectrum from established nutritional management practices to advanced pharmaceutical treatments and innovative microbiome-targeted approaches. Each strategy addresses a different component of the pathogenesis of ketosis, from limiting adipose tissue mobilization and supporting hepatic gluconeogenesis to reshaping microbial ecosystems. The integration of these interventions into comprehensive herd health programs, guided by precision monitoring and individualized risk assessment, represents the future of ketosis management [58].

By moving beyond reactive treatment toward proactive, multifaceted prevention, the dairy industry can reduce the incidence and impact of ketosis, improving both cow welfare and farm profitability (Figure 2).

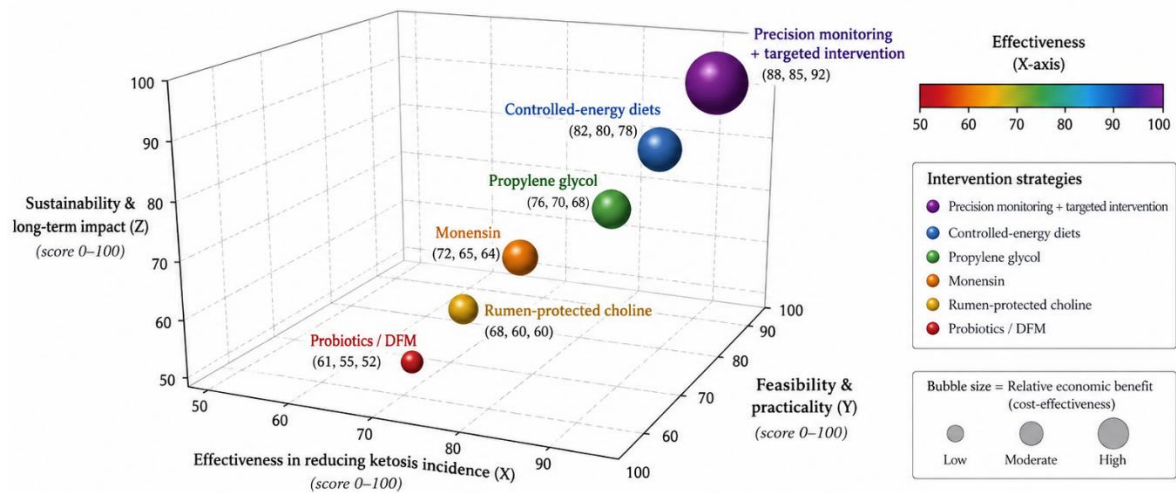


Figure 2. Comparative effectiveness of intervention strategies for ketosis prevention.

Each sphere represents a ketosis prevention strategy positioned according to its estimated effectiveness in reducing disease incidence (X-axis), feasibility of practical implementation at the farm level (Y-axis), and sustainability or long-term impact on animal health and productivity (Z-axis). Bubble size reflects relative cost-effectiveness, while spatial distribution allows simultaneous visualization of the multidimensional benefits and limitations of individual interventions. Scores range from 0 (lowest) to 100 (highest).

8. Wider impacts: reproduction, immunity, antimicrobial use and welfare

The consequences of ketosis in dairy cattle extend far beyond the immediate metabolic disturbance and temporary decline in milk yield [7,16,67,68]. Increasingly, research demonstrates that ketosis exerts systemic effects across multiple biological domains, shaping reproductive efficiency, immune competence, antimicrobial usage patterns, and overall welfare outcomes. These wider impacts are of particular concern in modern dairy systems, where the emphasis on high productivity coincides with societal and regulatory demands for sustainable, welfare-friendly production with minimal reliance on antimicrobials. Recognizing ketosis not only as a metabolic disease but as a multifaceted determinant of health and productivity is therefore crucial for advancing prevention and management strategies. Reproductive performance is one of the most consistently affected outcomes in cows experiencing ketosis. Elevated concentrations of circulating ketone bodies, particularly β -hydroxybutyrate (BHB), impair ovarian function through several mechanisms. Studies indicate that ketosis alters follicular fluid composition, reducing glucose and insulin-like growth factor availability while increasing inflammatory mediators, thereby compromising oocyte quality [7,16,67,68].

Luteal insufficiency, delayed resumption of ovarian cyclicity, and reduced conception rates are common sequelae [67-70]. Subclinical ketosis, although less overt than clinical disease, has been linked with prolonged days open, increased services per conception, and higher culling risk due to infertility. These reproductive consequences carry substantial economic costs, as the delay in returning cows to pregnancy extends calving intervals and decreases lifetime productivity. Recent advances in reproductive physiology also suggest that ketosis interacts with epigenetic regulation in the oocyte and early embryo, potentially influencing developmental competence in ways that extend beyond the immediate lactation. The impact of ketosis on immunity is equally profound, reflecting the intimate connection between metabolic status and immune function. During the transition

period, cows naturally experience a degree of immunosuppression, but ketosis amplifies this vulnerability. Elevated NEFA and BHB concentrations impair neutrophil chemotaxis, phagocytosis, and oxidative burst, reducing the cow's capacity to contain bacterial challenges in the uterus and mammary gland. This immunosuppressive environment explains the strong epidemiological association between ketosis and increased incidence of metritis and mastitis in early lactation. Moreover, chronic low-grade inflammation induced by adipose tissue mobilization and hepatic lipidosis further compromises adaptive immune responses, including lymphocyte proliferation and antibody production. The result is a dual burden of heightened susceptibility to infectious disease and reduced vaccine responsiveness, both of which complicate herd health management. The interplay between ketosis and immunity directly influences antimicrobial use patterns in dairy herds. Because ketotic cows are more likely to develop secondary diseases such as mastitis and metritis, antimicrobial treatment rates are correspondingly elevated. From a veterinary public health perspective, this link is critical, as dairy production is under increasing scrutiny for its contribution to antimicrobial resistance. Preventing ketosis therefore emerges not only as a strategy to improve cow health but also as a mechanism to reduce antimicrobial reliance in the dairy sector. The indirect pathway through which a metabolic disease drives antimicrobial demand underscores the importance of integrating metabolic health into antimicrobial stewardship initiatives. Farmers, veterinarians, and policymakers alike are recognizing that effective control of ketosis has implications that extend beyond the individual animal, contributing to global efforts to mitigate resistance development. Animal welfare considerations further broaden the impact of ketosis. Subclinical ketosis, often underdiagnosed, is nevertheless associated with reduced activity, altered feeding behavior, and decreased social interaction, all of which reflect compromised well-being. Clinical ketosis, with its overt depression, reduced appetite, and sometimes neurological manifestations, is even more clearly associated with suffering. Welfare is not limited to the avoidance of pain and illness but encompasses the ability of animals to thrive and express normal behaviors; in this context, ketosis undermines welfare on multiple fronts. Cows with ketosis often exhibit decreased lying comfort due to metabolic malaise, increased risk of lameness through altered energy balance, and higher culling probability, which prematurely terminates productive life. These outcomes conflict with both consumer expectations and emerging certification standards that emphasize welfare as a central component of sustainable livestock farming. Linking reproduction, immunity, antimicrobial use, and welfare reveals an interconnected cascade of impacts. A ketotic cow that fails to conceive in a timely manner not only contributes to reduced herd reproductive efficiency but may also experience repeated hormonal interventions and eventual culling, raising welfare concerns. Similarly, a cow with impaired immunity due to ketosis is more likely to require antibiotic treatment, raising both welfare (due to the disease episode itself) and antimicrobial stewardship issues. These interactions underscore the importance of holistic approaches to ketosis prevention: by intervening at the metabolic level, one simultaneously addresses downstream reproductive, infectious, and welfare challenges. Recent research has also highlighted the role of individual variability in shaping these wider impacts. Some cows appear more resilient to the reproductive and immune consequences of ketosis, suggesting genetic or epigenetic modifiers of susceptibility. Understanding these modifiers could help identify cows at greatest risk, allowing for targeted interventions that not only prevent ketosis but also protect against its cascading effects.

Similarly, studies of behavioral indicators reveal that [67-70] cows with subclinical ketosis exhibit subtle changes in rumination and feeding patterns before metabolic thresholds are reached, offering an opportunity to link welfare monitoring with early disease detection [34-36,71]. From a management perspective, the broader impacts of ketosis reinforce the need for integrated herd health programs. Reproductive management, antimicrobial stewardship, and welfare monitoring cannot be optimized in isolation from metabolic health. For example, aligning nutritional strategies with reproductive targets reduces both metabolic disease incidence and days open. Similarly, welfare assessments that include behavioral and metabolic indicators provide a more complete picture of cow well-being. By situating ketosis at the center of herd health planning, producers can align productivity goals with societal expectations for responsible antimicrobial use and animal welfare. Ketosis exerts far-reaching effects that extend beyond the immediate metabolic imbalance.

Its detrimental influence on reproductive function, immune competence, and disease susceptibility drives increased antimicrobial use, while its impact on behavior and culling risk compromises welfare outcomes. Addressing ketosis is therefore a multidimensional challenge with implications for farm profitability, public health, and ethical livestock production. Strategies that successfully mitigate ketosis will not only improve milk yield and metabolic stability but also enhance reproductive efficiency, reduce antibiotic dependency, and promote animal well-being. Recognizing and acting on these wider impacts is a defining step toward sustainable dairy production in the twenty-first century [34-36,71].

9. Economics, sustainability and adoption barriers of new technologies

The integration of advanced technologies for ketosis detection and management in dairy herds offers significant potential benefits but also presents economic and practical challenges that influence adoption [52-54,78]. Precision monitoring systems, metabolomic and lipidomic diagnostics, and microbiome-targeted interventions can improve early disease detection, optimize treatment, and reduce negative impacts on reproduction, immunity, and welfare. From an economic perspective, these technologies can increase milk yield, reduce veterinary costs, and decrease culling rates, resulting in improved profitability over the long term. Cost-benefit analyses consistently demonstrate that preventive interventions, particularly those enabling early identification of at-risk cows, are often more economical than reactive treatment strategies for clinical ketosis. Despite these potential returns, adoption remains constrained by several factors. Initial investment costs for sensor infrastructure, automated milking systems, and data management platforms can be prohibitive, particularly for small- and medium-sized farms. Maintenance, calibration, and training requirements further increase operational complexity. In addition, the interpretation of complex multi-omics data requires specialized expertise, limiting the practical application of these technologies outside research settings. Data integration and interoperability challenges among devices from different manufacturers can reduce efficiency and limit real-time decision-making, further delaying adoption. Sustainability considerations provide an additional incentive for technology adoption. By reducing the prevalence of ketosis and its associated complications, these interventions contribute to decreased antimicrobial use, lower greenhouse gas emissions per unit of milk through improved productivity, and enhanced animal welfare. Nutritional optimization and microbiome management can also reduce feed wastage and support more efficient nutrient utilization. In this context, technology adoption aligns with broader societal demands for environmentally responsible and welfare-friendly livestock production. Barriers to uptake are not purely economic. Farmer perceptions, risk aversion, and resistance to change influence the adoption of novel monitoring systems and interventions. Even when technologies demonstrate clear benefits in experimental trials, their perceived complexity or uncertainty regarding return on investment can limit real-world implementation.

Furthermore, regional regulatory frameworks and market structures may constrain access to pharmaceuticals, [52-54,78], supplements, or microbial-based products, affecting the feasibility of certain interventions [52,78-80]. Education, extension services, and demonstration farms are therefore critical to facilitate knowledge transfer and build confidence in emerging tools. While new technologies for ketosis management offer substantial economic, welfare, and sustainability benefits, adoption is moderated by financial, technical, and behavioral barriers. Overcoming these constraints requires strategies that integrate cost-effective solutions, user-friendly interfaces, and training programs to ensure practical implementation. By addressing these adoption barriers, the dairy industry can leverage innovations to enhance metabolic health, improve productivity, and promote sustainable and responsible farming practices [52,78-80].

10. Knowledge gaps and priority research directions

Despite substantial advances in understanding ketosis in dairy cattle, significant knowledge gaps remain that limit the development of fully effective prevention and management strategies [21,23,28,29]. One major gap lies in the integration of multi-omics data with practical herd-level outcomes. While metabolomics, lipidomics, and microbiome profiling have revealed novel biomarkers and mechanistic insights, the translation of these findings into predictive, actionable tools for farmers is still limited. Many identified biomarkers have been validated under controlled experimental conditions but require large-scale, longitudinal studies across diverse herds, diets, and management systems to ensure robustness and generalizability. Another critical area of uncertainty concerns the temporal dynamics of metabolic changes leading to ketosis. Although early detection is key to effective intervention, the precise sequence and interactions between adipose tissue mobilization, hepatic metabolism, rumen fermentation, and immune responses are not fully elucidated. Understanding these dynamics is essential for optimizing the timing and combination of nutritional, pharmaceutical, and microbiome-targeted interventions. Additionally, the individual variability among cows—shaped by genetics, epigenetics, and early-life nutrition—remains poorly characterized, limiting the ability to develop truly personalized prevention strategies. The role of the rumen microbiome in modulating host metabolism is increasingly recognized, yet mechanistic understanding of microbe–host interactions remains incomplete. Specific microbial taxa or metabolites that reliably predict ketosis risk are not consistently identified across studies, and the causative versus correlative nature of observed changes is often unclear. Further research is needed to define functional microbial networks, their interactions with host metabolism, and how these can be manipulated safely and effectively through diet, probiotics, or microbial transplantation. Precision monitoring technologies and sensor-based early detection tools present another area requiring further investigation. While promising, the optimal integration of multi-sensor data streams, development of standardized algorithms, and validation under commercial farming conditions are ongoing challenges. Questions remain regarding the cost-effectiveness, scalability, and farmer adoption of these technologies, particularly in smaller or resource-limited operations. Additionally, combining sensor data with metabolomic, lipidomic, or microbiome information to generate predictive models represents a frontier that has yet to be fully explored. Finally, the long-term impacts of ketosis on animal welfare, reproduction, and herd-level productivity require additional study. While associations between ketosis and reproductive inefficiency, immune suppression, and increased antimicrobial use are established, the mechanisms linking metabolic stress to these outcomes are not fully understood. Research addressing how interventions can simultaneously mitigate ketosis and improve welfare, fertility, and antimicrobial stewardship is urgently needed. In terms of priority research directions, several themes emerge. Longitudinal, multi-omics studies across diverse herd environments are essential to validate biomarkers and clarify causal pathways. Integrative studies combining metabolic, microbial, behavioral, and physiological data will enhance predictive modeling and support precision management approaches.

Mechanistic investigations into host–microbe interactions and their influence on lipid mobilization, hepatic function, and immune responses will help identify novel targets for intervention [21,23,28,29]. Finally, implementation research evaluating the economic, behavioral, and regulatory barriers to adoption of new technologies will be crucial to ensure that scientific advances translate into practical improvements in herd health, sustainability, and welfare [21,35,36,84]. Addressing these knowledge gaps will not only deepen understanding of ketosis pathophysiology but also enable the development of integrated, evidence-based strategies that reduce disease incidence, improve productivity, enhance animal welfare, and support sustainable dairy production systems [21,35,36,84].

Conflicts of Interest: The authors declare no conflicts of interest.

Declaration of Generative AI Use: The authors declare that no generative AI tools were used in the preparation of this manuscript.

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