



Organic nitrate 3-nitrooxypropanol (3-NOP) as a Methane-Reducing Feed Additive for Ruminants

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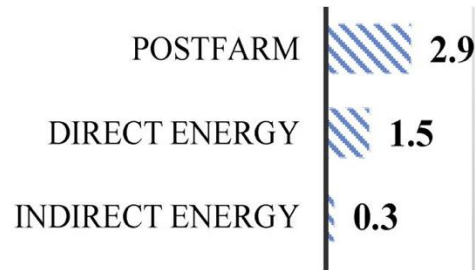
Section for Animal Health and Welfare

April 10th, 2026

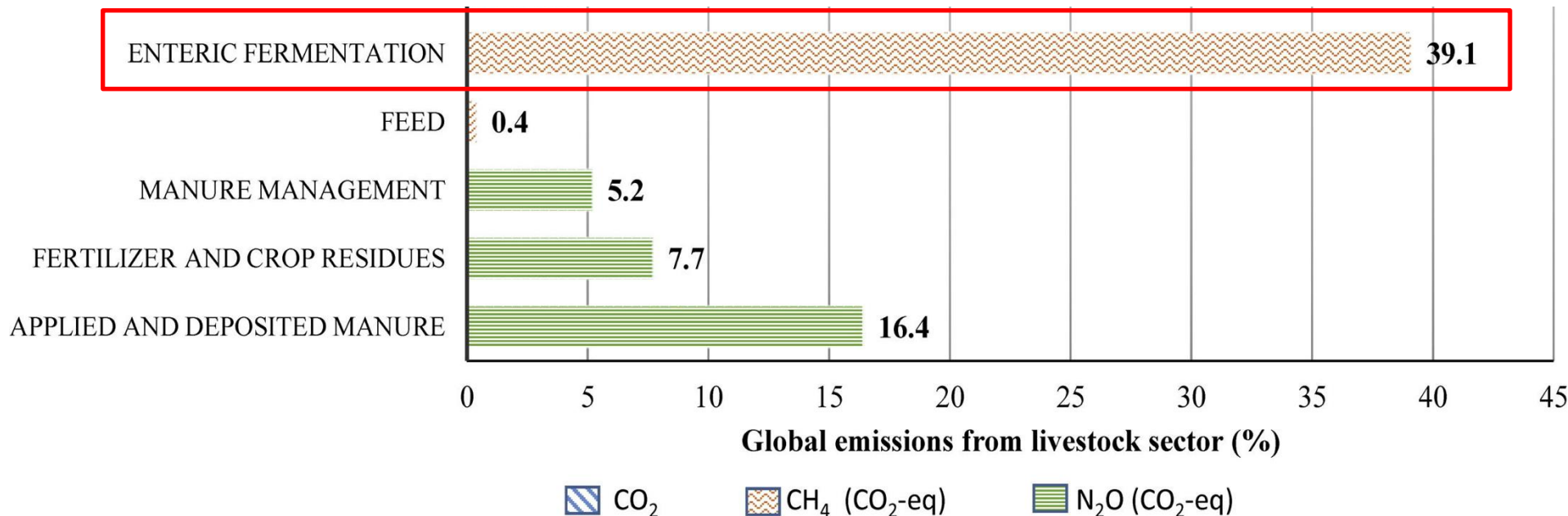
Structure of the Lecture

- Methane emissions in ruminants
- The rumen environment
- Strategies to mitigate enteric CH₄ emissions in ruminants
- 3-NOP
 - Mechanism of action
 - Degradation by-products
 - Effects on methane emissions
 - Current issues
- Conclusions

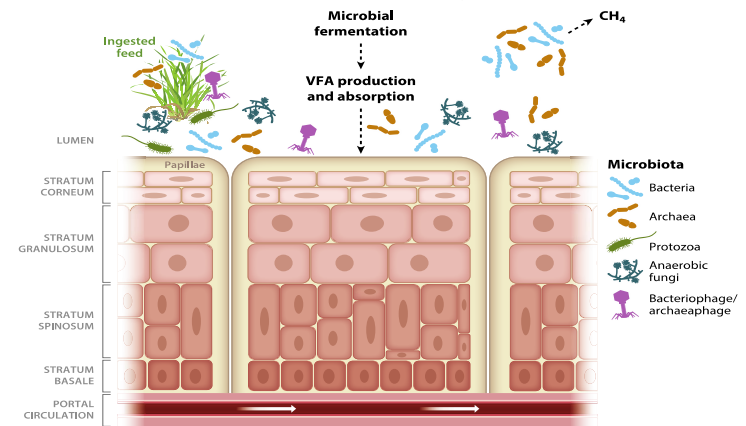
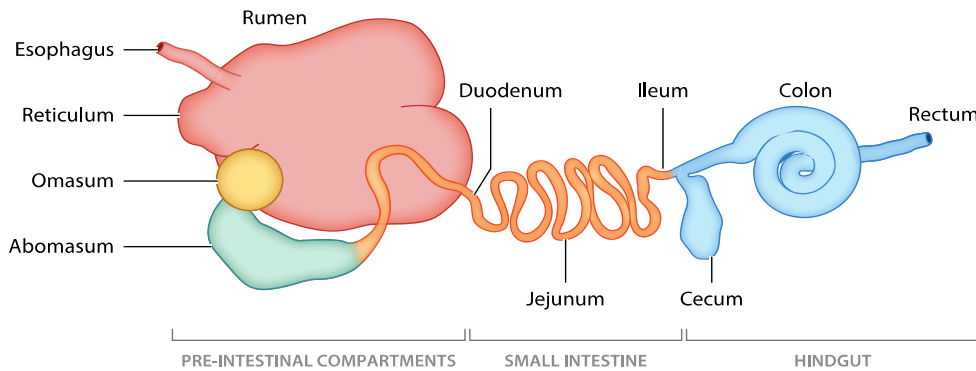
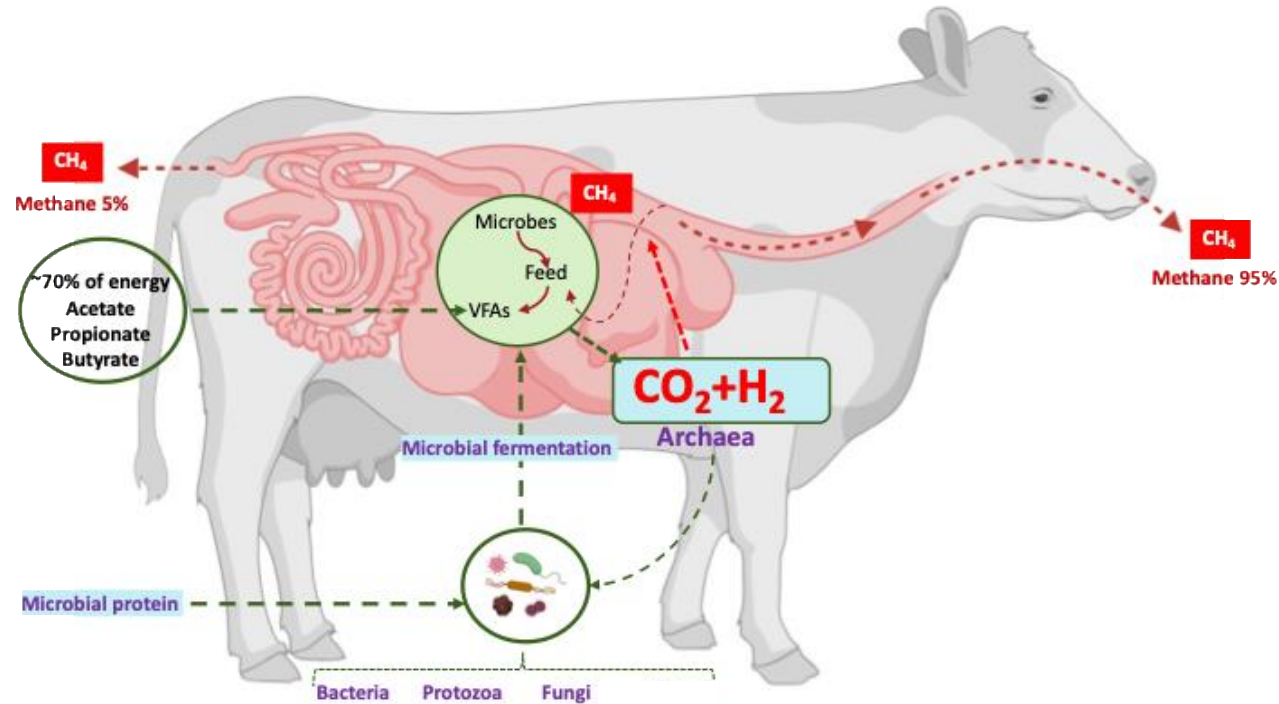
Enteric CH₄ reduction is needed



CH₄ emissions represent 2-12% energy loss from the gross energy intake in cattle (Johnson and Johnson, 1995)



Enteric Methane Production in Ruminants



Eóin O'Hara*, André L.A. Neves*, Yang Song, Le Luo Guan. 2020. The Role of the Gut Microbiome in Cattle Production and Health: Driver or Passenger?. *Annual Review Animal Biosciences*. 8:199-220.

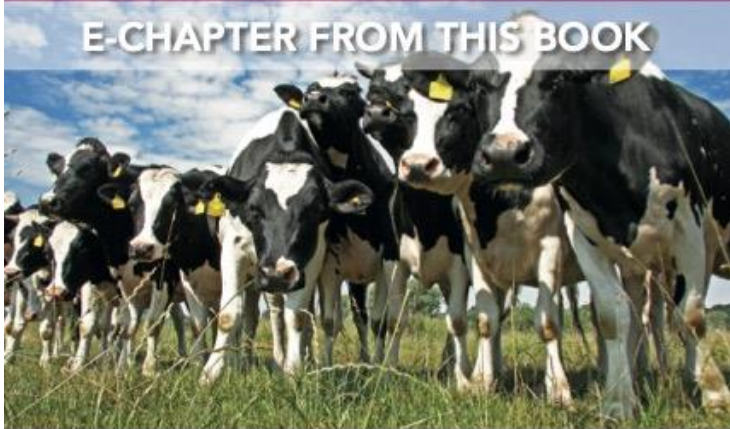
Strategies to reduce GHG emissions in ruminants

BURLEIGH DODDS SERIES IN AGRICULTURAL SCIENCE

Reducing greenhouse gas emissions from livestock production

Edited by Dr Richard Baines, Royal Agricultural University, UK

E-CHAPTER FROM THIS BOOK



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Modifying the rumen environment to reduce greenhouse gas emissions

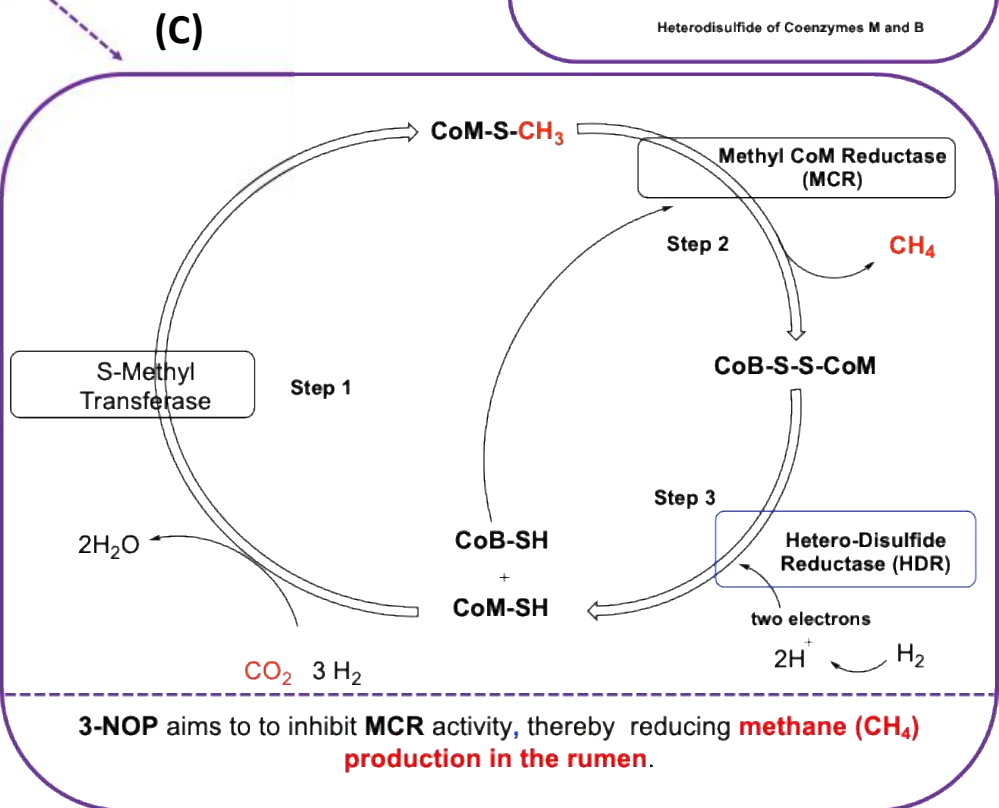
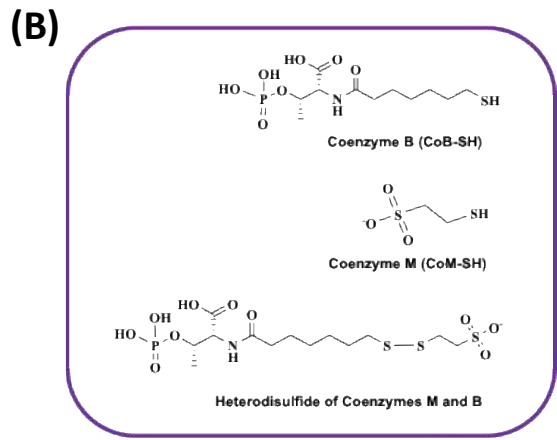
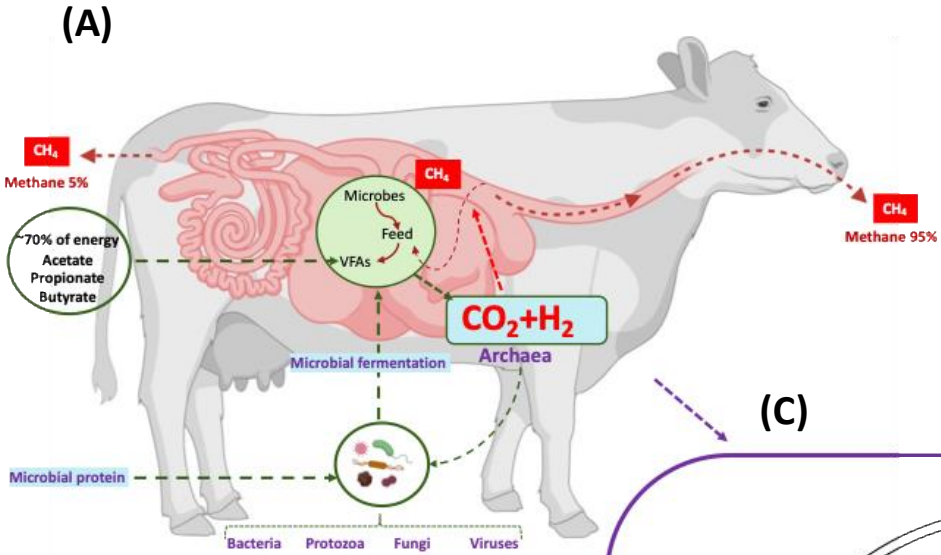
Yajing Ban, University of Alberta, Canada; André L. A. Neves, Embrapa Dairy Cattle, Brazilian Agricultural Research Corporation (Embrapa), Brazil; Le Luo Guan, University of Alberta, Canada; and Tim McAllister, Lethbridge Research and Development Centre, Agriculture and Agri-Food Canada, Canada

- 1 Introduction
- 2 Greenhouse gas production and the role of the rumen microbiome
- 3 Factors influencing methane production in ruminants
- 4 Modifying the rumen environment to reduce methane emissions
- 5 Conclusion
- 6 Where to look for further information
- 7 References

1 Introduction

The dramatic increase in the human population – estimated to reach 9.7 billion people by the year 2050 – will require an approximately 25% increase in gross agricultural output between 2020 and 2050 to meet the global food demand of humanity (FAO 2018; Nations 2019). However, increased animal production has placed an added strain on the environment as a result of the production of greenhouse gases (GHGs) (Huws et al. 2018) and nutrient accumulation in intensive livestock systems (Tullo et al. 2019). Ruminant production produces a number of GHGs including carbon dioxide (CO₂) mainly due to the use of fossil fuels, methane (CH₄) from enteric fermentation and manure, and nitrous oxide (N₂O) from manure and nitrogen fertilizer (Lynch and Pierrehumbert 2019). CH₄ is a particularly prominent GHG, as it has a global warming potential that is 28 times greater than that of CO₂ (Jackson et al. 2019). Globally, it has been shown that ruminants contribute about 11% of total anthropogenic GHG production, with approximately 6% arising from enteric CH₄ from ruminants (Rojas-Downing et al. 2017; Grossi et al. 2019; Beauchemin et al. 2020). In addition to the negative impacts on the environment, enteric CH₄ emissions can also represent a 2–12% loss in gross energy intake (Johnson and Johnson

Biochemistry of Methane Biosynthesis in the Rumen and Mitigation



Legend:

Step 1: S-Methyl Transferase methylates CoM-SH to give CoM-S-CH₃. A priori CO₂ and H₂ are consumed in the biosynthesis of the CH₃ group.

Step 2: MCR forms the heterodisulfide CoB-S-S-CoM from CoM-S-CH₃ and CoB, releasing methane.

Step 3: HDR reduces CoB-S-S-CoM to CoB-SH and CoM-SH.

Mechanism of action of 3-NOP

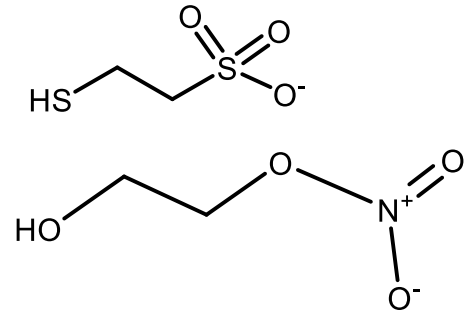


Mode of action uncovered for the specific reduction of methane emissions from ruminants by the small molecule 3-nitrooxypropanol

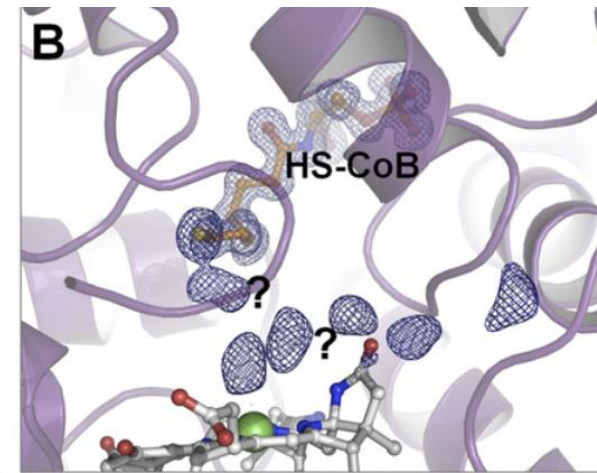
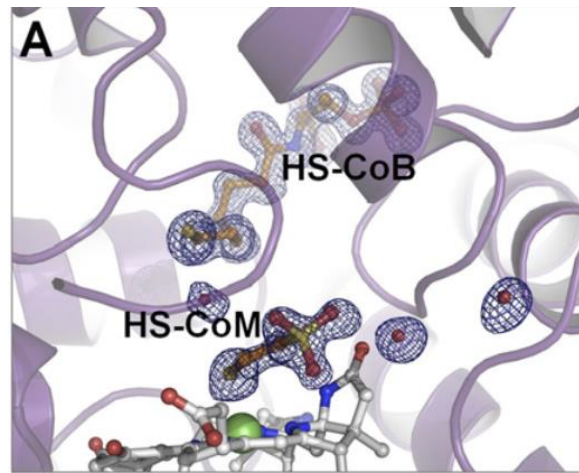
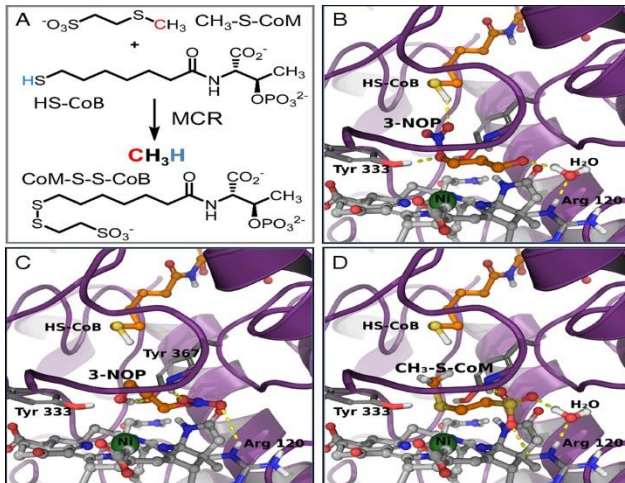
Evert C. Duin^a, Tristan Wagner^b, Seigo Shima^b, Divya Prakash^{b,1}, Bryan Cronin^a, David R. Yáñez-Ruiz^c, Stephane Duval^d, Robert Rümbeli^e, René T. Stemmler^e, Rudolf Kurt Thauer^{b,2}, and Maik Kindermann^{e,2}

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Structures of Coenzyme M (CoM) and 3-Nitrooxypropanol (3-NOP)



Target: methyl-coenzyme M reductase (MCR)



Metabolism of 3-NOP

- Mammalian metabolism:
 - 3-nitrooxypropanol (**3-NOP**) → 3-nitrooxypropionic acid (**NOPA**) → 3-hydroxypropionaldehyde (**3-HPA**)
 - **3-HPA** is the major urinary metabolite
- Rumen metabolism:
 - **3-NOP** → **propane-1,3-diol** (1,3-dipropanol is the major metabolite)
- In vivo observation
 - **NOPA** detected in milk ($\leq 3.66 \mu\text{g}/\text{kg}$) and in tissues ($\sim 5 \mu\text{g}/\text{kg}$)
- **NOPA** was not genotoxic

Bampidis et al., 2021. Safety and efficacy of a feed additive consisting of 3-nitrooxypropanol (Bovaer® 10) for ruminants for milk production and reproduction (DSM Nutritional Products Ltd). *EFSA Journal*, 19,

TABLE 1. Effect of 3-NOP (and other strategies) on enteric methane emissions

Item	Proportion of concentrate	Oilseed lipids (rapeseed and linseed)	Nitrate	3-NOP
Examined range in dietary inclusion	4 to 910 g concentrate/kg DM	21 to 85 g crude fat/kg DM	5 to 23 g nitrate/kg DM	28 to 200 mg 3-NOP/kg DM
Reported methane reduction (%)	22 to 41%	21 to 33%	6 to 28%	2 to 65%
Recommended supplementation level	100 g extra concentrate/kg DM	20 g extra crude fat/kg DM	10 g nitrate/kg DM	60 mg 3-NOP/kg DM
Potential methane reduction (%)	10%	7%	10%	30%

References related to 3-NOP research:

Haisan et al. (2014, 2017), Reynolds et al. (2014), Hristov et al. (2015), Lopes et al. (2016), van Wesemael et al. (2019), Melgar et al. (2020a, 2020c, 2021), van Gastelen et al. (2020; 2022), Schilde et al. (2021), Garcia et al. (2022), Kjeldsen et al. (2024), Maigaard et al. (2024).



Current Issues with 3-NOP (Bovaer®)

- **Large-scale rollout (Denmark):**
→ Adoption coincided with seasonal diet changes
- **Field reports (LVK; subset of herds):**
↓ rumination, ↓ feed & water intake, ↑ feed refusals, rumen atony, ↓ milk yield (~1–4 kg ECM), occasional sudden deaths
- **Emerging hypothesis: sulphur (S) interaction:**
3-NOP inhibits methanogenesis → ↑ H₂ → ↑ H₂S
(with high dietary S)



Conclusions

- **Methane mitigation with 3-NOP extends beyond MCR inhibition and requires a system-level understanding of rumen microbial chemistry and metabolic interactions**
 - **Key gap:** field conditions differ from controlled trials
- **EFSA (2026): call for additional data on the use of 3-NOP in ruminants**

Interested parties and stakeholders should submit data by **10/04/2026**, complying with the requirements described in the call.



Thank You for Listening
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