

New Approaches in Nutritional Management, Part 2

Reduced methane is helping the work of the rumen

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More ideas for reducing the environmental impact of beef cattle feeding. The optimisation of fermentation processes in the rumen can facilitate this objective. As, in this context, can the use of essential oils

As mentioned in the previous article, in addition to the use of former foodstuffs and co-products to increase the sustainability of the livestock industry, the environmental impact of meat production can be effectively optimised by supplementing nutrition with additives or foodstuffs that are able not just to optimise productive efficiency, but also to significantly reduce the production of methane.

This requirement has been essential to date, since the authorities involved in a Life Cycle Assessment (LCA) (Gold Standard) and those who issue carbon footprint reduction certificates, the Carbon Trust, now require that greenhouse gas emissions are clearly quantified, **especially** methane - the

greenhouse gas with the largest impact - as a result of its Global Warming Potential (GWP), which can reach values in the 25-28 range over a hundred-year period.

The Rumen - Specialist Surveillance

Indeed, Herrero et al (2013) have observed that methane, which by itself represents 32% of livestock emissions, is the main reason for environmental pollution attributable to cattle farming. Annually, the global rearing of ruminants leads to approximately 3.3 gigatons of CH₄, expressed as CO₂ equivalents, resulting primarily from cattle farming (beef and milk together 77%), followed by buffalo farming (13%), with smaller ruminants



accounting for the remainder (Fao 2019). And this is mainly attributable to the specific and intrinsic anatomical and physiological characteristics of ruminants, which lead to the production of significant amounts of methane in the rumen.

If, on the one hand, the complex ruminal biota, the specific environmental characteristics of the rumen, and their typical fermentative pathways enable ruminants to also convert fibre and fodder into nutrients, which in themselves are indigestible in monogastric animals, they do however, have a negative impact on the production of methane.

Indeed, the process of forming methane, methanogenesis, is in itself a tool for protecting the rumen: it allows metabolically-originated hydrogen, resulting from the metabolism of carbohydrates, and specifically from the re-oxidation of cofactors, that would constitute a risk to ruminal health and stability if released in the rumen to be inactivated and eliminated.

The hydrogen formed in this way is used by bacteria of the genus Archea and transformed into CH₄ to balance the pressure of hydrogen in the rumen, and is immediately emitted by eructation (Russel et al, 1997).

In addition to being a cause of environmental pollution, methane production also represents an energy loss for the animal, with a consequent reduction in productive efficiency. For these reasons, scientific research is focusing on the identification and development of strategies to optimise ruminal function, with a view to reducing methane emissions and concurrently increase efficiency in the conversion of nutrients.

The first system for modulating ruminal fermentation and reducing hydrogen production, and as a consequence methane, surely lies in the selection of raw materials to be included in feed. Depending on the source used, different fermentative pathways will be stimulated at ruminal level, characterised by varying degrees of efficiency and which have a different effect on the production of methane.

The acetate pathway, stimulated in the event of a high fibre content diet, is the least efficient from an energy perspective and releases a higher amount of hydrogen. On the other hand, the production of propionate, resulting from the greater use of cereals in feed, in addition to being more "efficient" from an energy point of view, requires hydrogen, therefore removing it from methanogenesis (Lozano et al, 2016, Mit-lhoener et al., 2013).

Figure 1 *In vitro* production of propionic acid and acetate (mg/l) after 16 and 24 hours of biomethanation

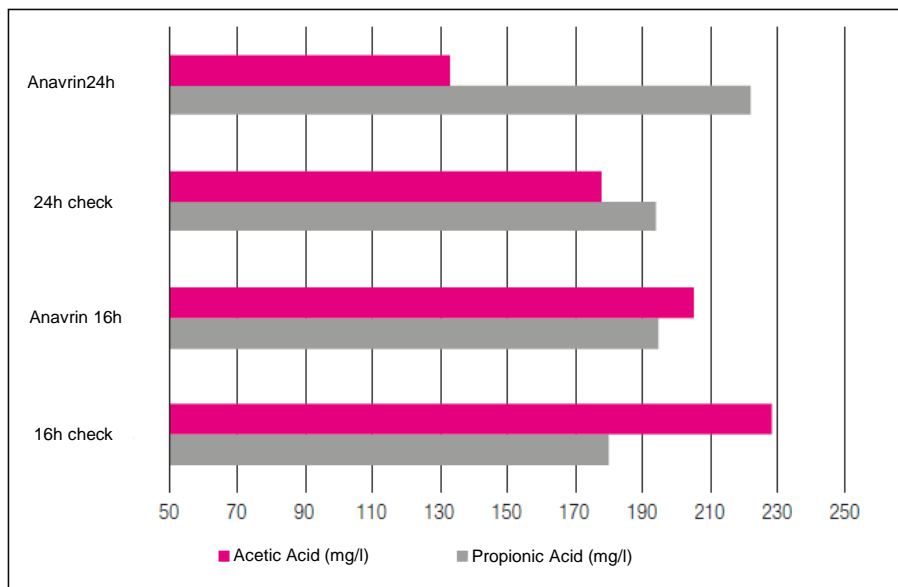
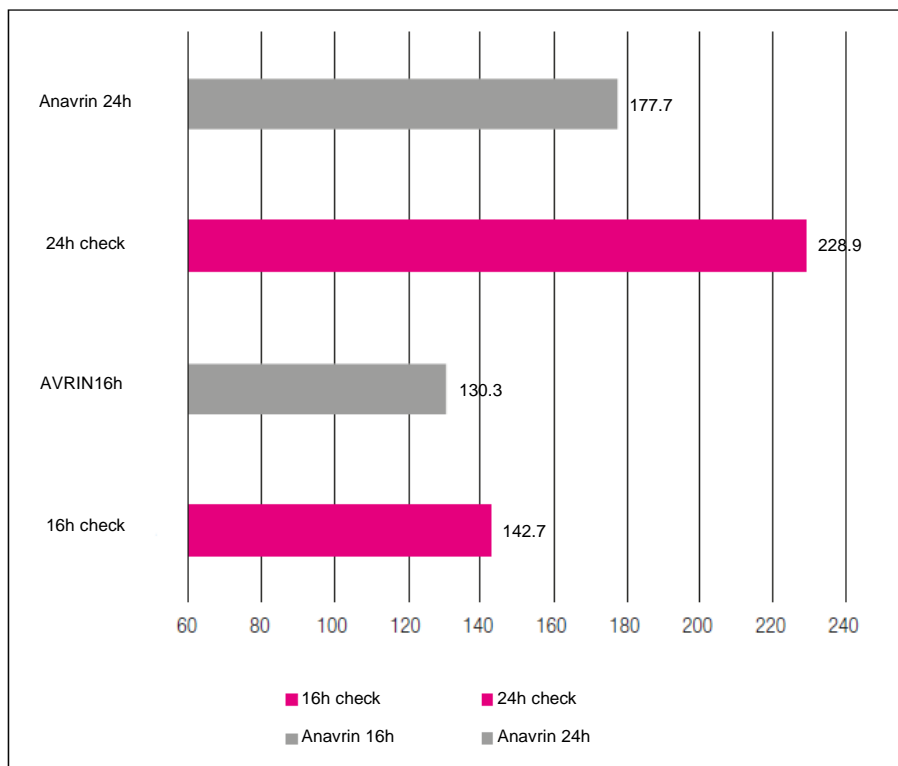


Figure 2 *In vitro* production of methane (BMPf) after 16 and 24 hours of biomethanation



Essential oils - a potential natural source of help

However, the choice of raw materials is

not the only viable means of modulating ruminal fermentations.

The production of CH₄ in the rumen can also be reduced by supplementing the

diet with specific additives that can inhibit the methanogenic microbial populations, causing either lysis or inactivation in them.

Table 1 Characteristics of software-rationing diets

| Method | Adaptation | Fattening |
|----------------------------------|------------|-----------|
| Corn silage | 8.00 | 8.00 |
| Corn flour | 2.50 | 5.50 |
| Wheat straw | 2.50 | 1.20 |
| Flour extracted from soya 44% PG | 0.70 | 1.00 |
| Husked sunflower 33% PG | 0.60 | 0.60 |
| Mineral and vitamin supplements | 0.17 | 0.20 |
| Unaltered, kg | 14.47 | 16.55 |
| Dried substitute, kg | 8.54 | 10.38 |
| Dried substitute, % | 59.10 | 62.74 |
| UFC, kg dried substitute | 0.85 | 1.02 |
| PG, % dried substitute | 11.80 | 14.28 |
| Soluble PG % dried substitute | 3.25 | 4.49 |
| Soluble PG, % of degradable PG | 44.21 | 48.96 |
| Sugars, % dried substitute | 4.32 | 4.15 |
| Starch, % dried substitute | 29.61 | 43.24 |
| NDF, % dried substitute | 43.55 | 29.20 |
| ADF, % dried substitute | 26.80 | 16.63 |
| ADL, % dried substitute | 4.91 | 3.25 |
| Lipids, % dried substitute | 2.54 | 2.95 |
| Ca, % dried substitute | 0.77 | 0.60 |
| P, % dried substitute | 0.29 | 0.32 |

In this regard, the use of natural substances like essential oils, bioflavonoids, and tannins, secondary metabolites from certain plants and spices that have an antimicrobial action, is also gaining increased ground in practice, given their specific properties, with the aim of achieving a reduction in methane production and,

concurrently, more efficient use of nutrients (Broudicou et al., 2000, 2002; Cardozo et al., 2004, 2005; Mohammed et al., 2004; Busquet et al., 2005, 2006, Calsamiglia et al., 2006). They are, in fact, capable of manipulating ruminal microbiota and of altering the ruminal fermentative pathways.

Firstly, the selection of specific ruminal bacterial populations causes an increase in the production of volatile fatty acids, specifically butyrate and propionate and also a reduction in the ruminal concentration of ammoniacal nitrogen, due to the lower protein degradation in this location, these are aspects that optimise the uptake of nutrients, growth and efficiency, simultaneously reducing environmental pollution.

Secondly, some *in vitro* studies have also effectively demonstrated how some oils derived from thyme, oregano, cinnamon, and garlic can reduce CH₄ production, exercising a direct effect on methanogens, with a further improvement on productive efficiency, as well as on environmental sustainability (Benchaar et al., 2011).

Essential oils also have an anti-inflammatory and anti-oxidising action which, by improving the physio-metabolic state of the subject, stimulate increased immune function, with positive effects on the health and the general condition of animals (Harborne and Williams, 2000; Reddy et al., 2003; Lee et al., 2003).

The Results of Testing in the Field

For this purpose, a scientific study was carried out aimed at assessing the effect of a pool of essential oils, bioflavonoids, and tannins (Anavrin) Vetos Europe Sagl, Cadenazzo TI Switzerland), on the health, immune function, and growth performance during the entire rearing period. This *in*

vivo study was also combined with an *in vitro* investigation, conducted with the support of biometanators, in which methane production was evaluated. The investigation involved 210 charolais beef cattle, housed in grids in a specialist beef cattle fattening facility in Veneto. The subjects were followed for the entire fattening period (182 days).

On arrival, all of the subjects were individually weighed and divided, balanced by weight and shape, between the two study groups, and fed a unifeed diet (Table 1) with the only difference being the supplements of the pool of Anavrin essential oils (Vetos Europe Sagl – Cadenazzo TI Switzerland), to the treated group:

- Check: 105 bullocks fed unifeed *ad libitum*.

Anavrin: 105 bullocks feed unifeed supplemented with Anavrin *ad libitum* in quantities of 5g/per head/per day, included directly in the mineral vitamin mix to optimise mixing.

The adaptation diet was limited to being administered in the first two weeks following arrival, after which, the fattening diet was distributed to the animals.

In addition to the 'traditional' indicators of productive efficiency, related to growth performance and general health in the two study groups, other indicators of immune function and, *in vitro*, methane production were assessed through blood tests.

A) Productive Performance

As shown in Table 2, the animals in the Anavrin group showed a significant improvement in growth performance over the course of the entire rearing period.

Specifically, this difference was equivalent to 80 g/per head/per day in the first 102 days following arrival, and 70 g/per head/per day when considering the entire rearing period.

Table 2 Productive Performance

| | No. | Weight d ₀ , kg | Weight d ₁₀₂ , kg | Weight d ₁₈₂ , kg | IPMG ₀₋₁₀₂ , kg/d | IPMG ₁₀₂₋₁₈₂ , kg/d | IPMG ₀₋₁₈₂ , kg/d | Consumption, kg ss/d | ICA |
|---------|-----|----------------------------|------------------------------|------------------------------|------------------------------|--------------------------------|------------------------------|----------------------|--------|
| Check | 105 | 417.84 | 572.69 | 709.82 | 1.52 | 1.71 | 1.60 | 11.18 | 7.04 |
| Anavrin | 105 | 416.37 | 579.81 | 720.86 | 1.60 | 1.76 | 1.67 | 10.56 | 6.37 |
| P | | ns | <0.05 | <0.05 | <0.001 | ns | <0.05 | <0.001 | <0.001 |

Table 3 Characteristics of the Carcasses

| | Carcass weight, kg | Yield, % | Conformation | | Fattening status | |
|----------|--------------------|----------|--------------|------|------------------|---------|
| | | | E, % | U, % | Cat. 2 % | Cat. 3% |
| Check | 418.31 | 58.92 | 95.24 | 4.76 | 56.19 | 43.81 |
| Anavrin | 425.87 | 59.07 | 96.19 | 3.81 | 59.05 | 40.95 |
| <i>P</i> | <0.001 | ns | Ns | | ns | |

Table 4 Health Status

| | Bovine Respiratory Syndrome (BRD) | | | | Lameness, % (n) |
|----------|-----------------------------------|-----------------------|-----------------------|---------------|-----------------|
| | First episode, % (n) | 1st recurrence, % (n) | 2nd recurrence, % (n) | Total, % (n) | |
| Check | 19.05 (20) | 8.57 (9) | 0.95 (1) | 28.57 (30) | 1.90 (2) |
| Anavrin | 13.33 (14) | 3.81 (4) | 0.00 (0) | 17.14 (18) | 1.90 (2) |
| <i>P</i> | 0.08 | 0.08 | Ns | <0.05 | Ns |

The best growth performance was achieved with a lower average quantity of feed consumption, reflecting the more efficient use of nutrients. This hypothesis is supported by a significantly improved feed conversion index in the group that was supplemented with the pool of Anavrin essential oils.

Furthermore, this supposed increased efficiency derived from dietary management with Anavrin was further witnessed in the *in vitro* analysis, which demonstrated a higher production of propionate, the most 'viable' volatile acid on the one hand, and a lower production of the least efficient acid acetate on the other, following the inclusion of percentages of product corresponding to the degree of inclusion in the actual diet, in relation to the control analysis (Figure 1).

Specifically, in the Green Plus group, the production of acetic acid was lower than 10% and 25%, whereas the production of propionic acid increased by 8% and 14% respectively after 16 and 24 hours of incubation. Additionally, an inhibiting effect on methanogenesis was also shown with Green Plus, another factor which may

explain the improved productive efficiency in the treated group. In particular, the production of methane (BPMf), still referring to the percentages corresponding to those used *in vivo*, led to a reduction of 9% and 22% of methane produced after 16 and 24 hours of incubation (Figure 2) with a spike of a 34% reduction, when included in greater quantities.

In relation to slaughtering performance, as shown in Table 3, with the exception of carcass weight, better in the Anavrin group as an effect of a higher live weight at the end of the fattening cycle, no significant differences emerged that could be attributed to the dietary management with regard to the yield, conformation and the state of the fattened carcasses.

B) Health Status

Proceeding to an analysis of the health-related data, the nutritional management seemed to have given the animals an improved capacity to adapt to new environmental conditions and to respond to stressful events affecting stored beef cattle, subject to lengthy travel, mixing, and radical social and environmental changes.

In terms of general health, the subjects



supplemented with the pool of Anavrin essential oils actually showed a significantly lower overall morbidity from respiratory diseases (Table 4, 28.57% v 17.14%), whereas no difference in instances of lameness emerged.

The positive effects on health following treatment may be justified in an aspecific increased immune response, and in a greater anti-oxidising reaction. Serum bactericidal (Table 5) 45 days after arrival was indeed better in the Anavrin group, which received the nutraceutical pool, significantly above the 90% threshold, a value believed to be the limit for healthy bovines (Amadori et al., 2002).

In relation to improved antioxidant capacity, and therefore the response to oxidative stress, OXY values at 45 days are significantly higher in the Anavrin group, in relation to the check group.

No difference was observed in the specific immune response (seroconversion of antibodies to the BHV-1 vaccination), gamma-interferon and ROMs, with values that were found to be overlapping.

Table 5 Blood Chemistry Parameters

| | Check | Anavrin | P |
|--|--------|---------|--------|
| BHV-1 serum neutralisation, log(dilution) | | | |
| d0 | 0.00 | 0.00 | ns |
| d45 | 0.78 | 0.78 | ns |
| Serum bactericidal, % | | | |
| d0 | 68.60 | 67.80 | ns |
| d45 | 80.40 | 92.00 | <0.001 |
| γ-interferon, pg/ml | | | |
| d0 | 15.00 | 15.60 | ns |
| d45 | 14.60 | 14.80 | ns |
| ROM, U/Carr | | | |
| d0 | 51.64 | 50.99 | ns |
| d45 | 68.62 | 68.39 | ns |
| OXY, μmol HClO/ml | | | |
| d0 | 257.52 | 255.30 | ns |
| d45 | 290.58 | 356.68 | <0.001 |

Conclusions

The analysis of the results that emerged from this study lead us to believe that the best growth performances observed are due to the optimisation of the fermentation processes in the rumen and the improved overall health following the use of the pool of Anavrin essential oils. These oils act by modulating the kinetics of ruminal fermentation, reducing the production of methane and encouraging that of fatty acids that are useful in terms of energy, but also by improving the health of animals that undergo stress as a result of their antioxidant activity and by stimulating immune response. The results of this study confirm these theories. •

Therefore, the results seem to confirm that certain essential oils are capable, in addition to modulating bacterial activity, of also improving health, thanks to their anti-inflammatory and immune response

stimulating action (Harborne and Williams, 2000; Reddy et al., 2003; Trouillas et al., 2003; Guetiérrez et al., 2003, Lee et al., 2003).

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