REVIEW ARTICLE



Impact of essential oils on methane emissions, milk yield, and feed efficiency and resulting influence on the carbon footprint of dairy production systems

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Abstract

Reducing CO_2 emissions is one of the highest priorities in animal production. Regarding methane reduction, feed additives are of growing importance. As shown in a meta-analysis, the use of the essential oil (EO) blend Agolin Ruminant affects methane production per day (-8.8%), milk yield (+4.1%), and feed efficiency (+4.4%). Building on these results, the present study investigated the effect of varying individual parameters on the carbon footprint of milk. The environmental and operational management system REPRO was applied to calculate the CO_2 emissions. Calculation of CO_2 emissions include enteric and storage-related CH_4 , storage-, and pasture-related N_2O as well as direct and indirect energy expenditures. Three feed rations were created, differing in their basic feed components such as grass silage, corn silage, and pasture. Each feed ration was differentiated into three variants: variant 1 CON (no additive), variant 2 EO, and variant 3 (15% reduction of enteric methane compared to CON). Due to the reducing effect of EO on enteric methane production, a reduction potential of up to 6% could be calculated for all rations. Considering other variable parameters, such as the positive effects on ECM yield and feed efficiency, a GHG reduction potential of up to 10% can be achieved for the silage rations and almost 9% for the pasture ration. Modeling showed that indirect methane reduction strategies are important contributors to environmental impacts. Reduction of enteric methane emissions is fundamental, as they account for the largest share of GHG emissions from dairy production.

Keywords Essential oil \cdot CO₂ \cdot Carbon footprint \cdot Methane emission \cdot Dairy \cdot REPRO

Introduction

In 2050, the goal of the European Commission is to reach climate neutrality. The climate agreement of the UN Climate Change Conference in 2015 states that all sectors need to be

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participate in reducing GHG emissions (FAO 2019). Different strategies for reducing greenhouse gas production (GHG) emissions are widely discussed. The agricultural sector contributes 11% of global GHG emissions (OECD/FAO 2020). Nevertheless, the demand for livestock products is expected to double by 2050 (FAO 2019). Dairy production as a part of the livestock sector contributes 2.7% to total anthropogenic emissions (Gerber et al. 2011) and produced 852 million tonnes or 81% of global cow milk in 2019.

Due to an increased consumer demand to dairy products, overall milk production has increased by 30% and global dairy herds have grown by 11% between 2005 and 2015 (FAO 2019). Despite this, more efficient production methods have been applied on dairy and cattle farms for many years, resulting in a decreasing emission intensity per unit of product. According to FAO (2019), the GHG per kg of milk have declined by 11% between 2005 and 2015.

Adversely, milk production plays a major role in GHG emissions. The most anthropogenic gas is methane which is released through cow belches and manure, followed by carbon dioxide. Methane is a powerful gas which has a significant effect on the climate and therefore contributes to the tropospheric ozone formation (European Commission 2020). It has a short lifetime in the atmosphere of approximately 9–12 years, but CH₄ is 25 times more harmful than carbon dioxide (Nazaries et al. 2013). Nevertheless, it plays an important role in the biogenic carbon cycle to remove CO₂ from the atmosphere and recycle the carbon which is stored in plants and consumed by ruminants (Werth 2020). Livestock production contributes to 37% of global methane emissions (FAO 2006). About 80.7% are from enteric fermentation in ruminants (European Commission 2020).

Decreasing the enteric fermentation of methane in the rumen is a worldwide challenge for animal nutritionists and other scientists relating to microbiology or biochemistry. One strategy to reduce the release of methane from cattle is the manipulation of the microbial rumen ecosystem and fermentation kinetics to yield a higher feed utilization. Production efficiency diminishes factors like feed and emissions of GHG to produce a given quantity of output (e.g., milk). Increasing milk yields per cow are related to a lower input of feed intake to produce 1 kg of energy-corrected milk (ECM) as well as a reduction in the release of GHG per kg milk (FAO 2019).

Different strategies to reduce enteric methane production are widely discussed. Much recent scientific research relating to the use of essential oils and plant substances has been carried out. Due to their antimicrobial properties, EOs have been shown to have a specific effect on rumen bacteria related to altered fermentation in the rumen (Carrazco et al. 2020). The changes in rumen microflora could result in a reduced enteric methane production. Plant actives are able to inhibit protozoa (Patra and Yu 2012), impact on electron pathways, and affect the integrity of bacteria cell membranes, resulting in an influence on methanogenesis (Calsamiglia et al. 2007).

A reduction of methane release from the cow to improve the carbon footprint of milk is the focus of the current study. Three different feeding strategies were compared using the environmental and operational management system REPRO with the associated animal module. Database information is used from the meta-analysis carried out by Belanche et al. (2020a, b).

Materials and methods

Scope

Data used and system boundary

GHGs were calculated using data from the studies included in the meta-analysis by Belanche et al. (2020a, b). In the context of the study, the REPRO model was chosen as the basis for the GHG calculations because it is a modular agricultural software. A detailed explanation of the tool can be found in the paragraph tool description and carbon dioxide methodology. Thus, the contents to be processed can be adapted to the specific questions of this study and calculated via the three different scenarios. The system boundary includes all relevant processes from feeding through to the storage of the manure produced as indicated in Fig. 1.

The production of input quantities such as animals, husbandry system, pasture, and manure management are called foreground data. Background data contains all relevant inputs to the foreground system like electricity and water production as well as feed and indirect inputs (such as building materials used). To represent the foreground data, the input data are taken from the meta-analysis, such as milk



Fig. 1 System boundaries, subcategories, operating resources and emission sources

yield, milk fat, and protein content. The feed rations are related to the data of the meta-analysis. The imported feed was considered as input of nitrogen and phosphorus for the animal husbandry, but the emissions related to their production as well as emissions resulting from the construction of the husbandry system and the manure store were excluded. On the other hand, important production inputs, such as upstream energy use, were included. The sale of animals, milk, and manure as an output process can be included in the calculation; for this project, these aspects are not relevant. The model-based calculation of greenhouse gases focuses on the physiological and nutritional emissions of the dairy cow. Thus, no emissions of CO_2 from feed production, milk production, barn construction, and housing system are considered.

Formel 1

Allocation

In the current study, allocation of emissions between milk, surplus calves, culled cows, and manure was not conducted. All energy inputs and greenhouse gas flows were allocated to the ECM (kg/y) produced.

Functional unit

The functional unit of this study was 1 kg of energy-corrected milk (ECM). ECM was standardized for 4.0% fat and 3.3% protein content (Spiekers and Potthast 2004).

ECM (kg) = $((0.38 \cdot \text{milk fat}(\%) + \text{milk protein}(\%) + 1.05) \cdot \text{milk (kg)}) \div 3.28$

Impact assessment

The greenhouse gas emissions were determined using the current emission factors, which assume the global warming potential of CH_4 and N_2O for a 100-year time horizon. For CH_4 and N_2O , it was assumed to be 25 and 298 CO_2 equivalents respectively (IPCC 2007).

Performance data

The animal-related input data on milk yield and milk ingredients are taken from the metadata analysis Belanche et al. (2020a, b). In order to ensure the comparability of the results with other studies, the amount of ECM specified there was used as an initial value for further calculation. The ECM (32.9 kg/d) corresponds to the mean value of the ECM from the published studies. Based on the ECM and the non-standardized proportion of fat (4.03%) and protein (3.25%), the mean daily milk yield (33.1 kg/d) was recalculated (Spiekers and Potthast 2004).

Further animal-related data, such as liveweight and indicators of lactation, were taken in a standardized manner from the master data of the animal module. The lactating milk cow with a live weight of 650 kg was chosen as the group of animals. A lactating period of 305 days was scheduled between the start of lactation and the end of lactation. A dry period of 60 days duration was assumed.

Rations and variants

The determination of feed rations was based on the energy and protein requirements of a dairy cow producing 32.9 kg ECM. Initially, three feed rations were created, which differ in their basic feed components such as grass silage (ration 1), maize silage (ration 2), and pasture grass (ration 3). Each feed ration was differentiated into three variants: variant 1 CON (no additive), variant 2 EO (Agolin Ruminant additive), and variant 3 (reduction of enteric methane by 15% compared to CON). In feed ration 3, a daily grazing time of 8 h for 180 days was specified. As a result, nine different scenarios were calculated, which are shown in Table 1.

Variant CON and variant 15%-CH₄ reduction receive a milk yield of 33.1 kg per day at 10,084 kg per year annual output. Variant 2 EO is assigned a higher milk yield of 34.6 kg per day and an annual yield of 10,549 kg per year, which express the positive effect of EO on ECM yield (+4.1%) and feed conversion (FCE+4.4%).

The change in milk yield caused by the addition of EO resulted in an increase in yield from 10,084 to 10,549 kg/y. The additional consideration of feed efficiency resulted for variant 2 with EO additive in a final ECM performance of 10,497 kg/y.

Considering the defined milk yield of 33.1 kg/d, the protein and energy requirements of the dairy cow were calculated. The preparation of a TMR (total mixed ration) over the entire lactation period was assumed. Table 2 lists the feed rations

Milk yield		Feed ration										
		Ration 1			Ration 2			Ration 3				
		CON ^a	EO ^b	-15% CH ₄	CON	EO	-15% CH ₄	CON	EO	– 15% CH ₄		
Milk yield	kg/d	33.1	34.6	33.1	33.1	34.6	33.1	33.1	34.6	33.1		
	kg/y	10,084	10,549	10,084	10,084	10,549	10,084	10,084	10,549	10,084		
ECM ^c yield	kg/d	32.9	34.4	32.9	32.9	34.4	32.9	32.9	34.4	32.9		
	kg/y	10,035	10,497	10,035	10,035	10,497	10,035	10,035	10,497	10,035		

Table 1 Daily and annual amounts of milk and ECM in kilograms within the selected feed ration types and associated variants based on Belanche et al. (2020a, b)

^aCON, control variant

^bEO, essential oils blend additive

^c*ECM*, energy-corrected milk

Table 2 Components of the feed rations

Components of feed (%)	Feed ration								
	Ration 1	Ration 2	Ration 3						
			Summer	Winter					
Gras silage	46.1	31.6	-	-					
Maize silage	32.3	47.4	-	64.8					
Pasture grass	-	-	68.5	-					
Alfalfa hay	-	-	9.8	5.8					
Alfalfa silage	-	-	9.8	11.6					
Soybean meal (44% CP ^a)	5.1	4.5	-	4.6					
Winter wheat	7.6	7.4	3.4	3.5					
Grain maize	6.0	6.3	4.9	6.5					
Dried pulp	2.3	2.3	3.3	3.0					
Mineral feed	0.6	0.6	0.4	0.2					

^a*CP*, crude protein

used in the study. The feed values are stored in the master data for the calculations of the ration contents. They are taken from the feed database of the DLG—Deutsche Landwirtschaftsgesellschaft e. V. (German Agricultural Society). The data on ration composition are presented in Table 2.

The rations differ in their basic feed components, energy and nutrient contents. Ration 1 is grass silage-emphasized and has a share of 46.1%. Ration 2 is a maize silage-emphasized ration with a share of 47.4%. The third variant is a grazing ration with summer and winter feeding. The summer feed contains 68.5% pasture grass and 9.8% alfalfa hay. In contrast, the winter feed is focused on maize silage at 64.8% and alfalfa hay at 5.8%.

Feed intake (DMI—dry matter intake) was calculated using the model of Menke (1987). Live weight was assumed to be 650 kg, while milk yields were obtained from the metaanalysis compare Variant CON and variant 15%-CH₄ reduction receive a milk yield of 33.1 kg per day at 10,084 kg year Table 3 Inventory data for rations 13 normalized per day, per year, and to 1 kg dry matter

Feed values		Feed ration					
		Ration 1	Ration 2	Ration 3			
Dry matter intake	(kg DM/d)	20.0	19.8	20.2			
Energy	(MJ NEL ^a /kg DM ^b)	7.1	7.2	6.9			
Crude protein	(g/kg DM)	159.1	148.6	142.6			
Usable crude protein	(g/kg DM)	158.9	159.3	150.0			
Crude fat	(g/kg DM)	33.0	32.3	30.1			
Crude fiber	(g/kg DM)	154.6	144.5	188.0			
N-free extract	(g/kg DM)	577.0	606.1	567.2			
Crude ash	(g/kg DM)	63.8	56.6	64.2			
Ruminal N bal- ance	(g N/kg DM)	0.66	-0.85	-0.42			
N intake	(kg N/y)	197.8	184.4	180.5			
N excretion	(kg N/y)	129.8	119.2	115.3			
Volatile solids	(kg/d)	4.2	4.2	4.8			

^a*NEL*, net energy content for lactation

^bDM, dry matter

annual output. Variant 2 EO is assigned a higher milk yield of 34.6 kg per day and an annual yield of 10,549 kg per year, which expresses the positive effect of EO on ECM yield (+4.1%) and feed conversion (FCE + 4.4\%).

Calculated DMI for ration 1 is 20.0 kg DM/d, and for ration 2 this resulted in 19.8 kg DM/d. The total DMI for grazing ration was calculated based on how much feed intake must occur in the barn at a given amount of milk and maintain an animal at a given weight. For the grazing ration, a dry matter intake of 20.2 kg DM/d was calculated. The calculation of DMI in this model includes a 7% waste of feed (Spiekers et al. 2011). Data on feed intake, nutrient composition, and ruminal nitrogen balance (RNB) are presented in Table 3.

The usable protein at the duodenum consists of two components, the feed protein and the microbial protein. whereby the amount of microbial protein formed correlates closely with the energy intake. If the rumen microbes are supplied with sufficient energy, the protein supplied by the feed is converted into highly digestible "microbial protein." The protein requirement of ruminants is usually higher than the amount of microbial protein available at the duodenum. The difference must therefore be provided by non-microbially degraded feed protein. The feed values for the usable crude protein were taken from the DLG feed value tables (2013 ff.). The calculation excretion of volatile solids is mentioned in Table 4. The RNB represents an offset between crude protein and usable protein. It indicates whether there is a nitrogen surplus or deficiency in the rumen due to the corresponding feed. The RNB is calculated as follows:

Equation 2

$$\text{RNB} = \frac{\text{XP} - \text{nXP}}{6,25}$$

All three rations have an approximately balanced RNB. In the case of ration 1 with grass silage as a basic feed component, the RNB value is slightly positive, whereas rations 2 (maize) and 3 (pasture) are slightly negative.

Essential oil

Agolin® Ruminant (Agolin SA, Bière, Switzerland) is a commercially available blend of essential oils, containing coriander seed oil, eugenol, and geranyl acetate, which has been demonstrated to reduce greenhouse gas emissions in dairy cows and improve energy-corrected milk and feed efficiency (Elcoso et al. 2019).

Basis data of the husbandry system

During lactation, cows are kept in an individual housing system and the excrements are collected in specially designed storage facilities. This study does not represent a complex barn with connected manure storage, but key aspects of a modeled dairy farm system. The calculation of CO_2 emissions requires certain housing parameters, such as the stable

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Gas	Emission factor/equation	Methodology reference
CH ₄		
Enteric fermentation	Based on ration composition	RÖSEMANN et al. (2013), KIRCHGESSNER et al. (1994)
Manure storage	Slurry-based system	Tier 2 IPCC (2006)
Grazing	$VS^a = feed intake^b * (1 - X_{DOM}^c) * (1 - X_{ash, feed}^d)$	Dämmgen et al. (2011)
	$B_0^{e} = 0.23 \text{ m}^3 \text{ of kg VS}$	Dämmgen et al. (2012)
	$MCF_{Slurry}^{f} = 0.17 \text{ m}^{3} CH_{4} \text{ of } \text{m}^{3} CH_{4}$	Dämmgen et al. (2012)
	MCF $_{\text{Grazing}} = 0.01 \text{ m}^3 \text{ CH}_4 \text{ of } \text{m}^3 \text{ CH}_4$	HAENEL et al. (2020)
	DMI (kg/d)	MENKE (1987)
N ₂ O direct		
Manure storage	EF _{N2O-N direct, Slurry} =0.000 kg N ₂ O-N of N _{Slurry}	Tier 2 IPCC (2006)
Grazing	EF _{N2O-N direct, Grazing} =0.02 kg N ₂ O-N of N _{Grazing}	IPCC (2006)
N ₂ O indirect ^g		
House	$EF_{N2O-N indirect, House} = 0.197 \text{ kg NH}_3\text{-N of kg N}_{TAN, House}$	DÖHLER et al. (2002) and
		Dämmgen et al. (2010)
Grazing	$EF_{N2O-N indirect, Grazing} = 0.1 \text{ kg NH}_3\text{-N of kg N}_{TAN, Grazing}$	EMEP (2013)
Storage	EF _{N2O-N indirect, Slurry} =0.150 kg NH ₃ -N of N _{TAN, slurry}	DÖHLER et al. (2002) and
		Dämmgen et al. (2010)

Table 4 Detailed parameters, equations, and emission factors (EF) of enteric fermentation, husbandry system, and manure storage relevant to calculate the CH_4 , NH_3 , and N_2O emissions

^aExcretion of volatile solids (in kg/y)

^bFeed intake rate (dry matter) (in kg/y)

^cDigestibility of organic matter (in kg/kg)

^dAsh content of feed (in kg/kg)

^eMaximum methane producing capacity (in m³ CH₄/kg VS)

^fMethane conversion factor (in m³/m³)

^gVolatilisation of ammonia in the barn and manure storage which result in indirect emissions of nitrous oxide

type, the husbandry system, and the floor type. Therefore, a free stall housing system with a theoretical partially slatted floor area of 1 m^2 was assumed. Furthermore, an open slurry tank with a size of 1 m^3 was chosen for the fertilizers produced in the model. Other equipment such as drinking trough, feeding strategy as well as feed preparation and watering, cubicle housing, manure removal, and milking system were not considered.

Tool description and carbon footprint methodology

Tool description

As previously mentioned, the GHG emissions associated with the dairy production on-farm level were determined using the model REPRO. The REPRO model (reproduction of soil fertility, specifically organic matter) was developed in the 1990s at the Martin Luther University Halle Wittenberg (MLU) and the Technical University of Munich (TUM). It offers the possibility to assess the current state of agricultural crop production and animal husbandry with regard to its material and energy flows. Farms are understood as systems to which system boundaries and subsystems are assigned. The modeling implemented in REPRO offers the possibility to analyze the interactions across the system boundaries and between the subsystems on the basis of networked material and energy flows (Hülsbergen 2003). For the complex assessment of agricultural systems, it is necessary to integrate various partial aspects into one overall statement. This has the advantage over simple indicator approaches that the interactions between the system components can be represented. Furthermore, the connection between agricultural activities and environmental impacts can be worked out and, due to the scenario capability, planned changes in farming practices can also be presented. The model is supported in all modeling work by a comprehensive data pool of scientific-ecological parameters and systems of equations. The model is divided into 4 modules: Location, Crop Production, Animal Husbandry, and Storage. For each module, data processing follows the same principle of data collection, analysis, and evaluation (Christen et al. 2009). The program structure is modular and is basically divided into three work areas: mapping of the management system, analysis and calculation of indicators, and evaluation and presentation of results (Fig. 2).

Animal husbandry and the calculation of specific indicators are carried out in a separate module complex of REPRO (Becker et al. 2015). The coupling via feed and manure storage enables to represent all internal nutrient cycles between plant cultivation and animal husbandry. Data on animal husbandry are recorded and managed on a stable area basis, classified according to animal species and production directions, age classes, and performance groups. The basis is provided by operational herd data, performance tests, ration design, documents on stable construction as well as on-site surveys and



Fig. 2 Structure of the REPRO model and networking of the module

assessments. The feed requirement is calculated depending on the performance of the pasture or stable management. Depending on feeding, the amount and content of organic manure is calculated. Building on the information gathered, the analysis subsystem calculates the effects of the operation on the abiotic and biotic environment. Here, the material and energy cycles are balanced, relevant loss paths quantified, and intensities identified. The result are agri-environmental indicators that describe the overall system and have a high ecological relevance.

Carbon footprint methodology

GHG emissions quantified in the analysis include methane (CH_4) emissions from enteric fermentation and manure, ammonia (NH_3) emissions from barn and manure, nitrous oxide (N_2O) emissions from manure, and the energy intensity from direct energy use. The sums of the sub-indicators are expressed in CO₂ equivalents to take into account the different contributions of the gases to the greenhouse effect and their fate in the atmosphere recommended by (IPCC 2006a). Thus, the 25 times greater climate impact of N_2O are considered in the specified period of 100 years compared to that of CO₂.

Detailed parameters, equations, and emission factors (EF) of the husbandry system, manure storage, and grazing relevant to calculate the CH_4 , NH_3 , and N_2O emissions in this study are presented in Table 4.

Enteric methane emissions of dairy cows are calculated according to the guidelines of Rösemann et al. (2013) on the basis of the ration composition according to Kirchgessner et al. (1994). The decisive variables here are DMI and the relationships between crude fibre, N-free extracts, crude protein, and crude lipid content.

Methane production associated with storage was calculated using the tier 2 methodology according to IPCC (2006). Therefore, an EF was calculated as a function of volatile solids (VS) excreted, the maximum methane production capacity (BO), and the methane conversion factor (MCF), which indicates for each manure storage system the fraction of BO that is generated effectively. In this calculation, a MCF of 17% for slurry and 1% for manure dropped during grazing was used. VS excreted is calculated as a function of feed intake rate, digestibility of organic matter, and ash content of feed, derived by Dämmgen et al. (2011). Per cow, VS resulted for ration 1 and ration 2 in 4.2 kg VS/d and for ration 3 in 4.8 kg VS/d.

Direct N₂O emissions during storage and from soils during grazing were estimated using the IPCC (2006) default EF and depend on the nitrogen content of manure. To calculate nitrogen excretion, the total amount of N in feed intake minus the amount of N in milk and animals N was calculated. Per cow, this resulted for ration 1 in 197.8 kg N/y, for ration 2 in 184.4 kg N/y, and for ration 3 in 180.5 kg N/y (see Table 3). Total excretion is divided into organic N (excreted with feces) and TAN (total ammoniacal nitrogen, excreted as urine). The total excreta (organic N, TAN) are multiplied by the time shares in which the animals spend in the stable or on the pasture. In slurry systems, direct N₂O emissions are not assumed to occur there, as the emission factor is 0.00 kg N₂O-N/kg N. Direct nitrous oxide emissions from soils due to grazing the specific EF of 0.02 kg N₂O-N/kg N (IPCC 2006) was used. The calculated amount of N₂O-N is converted to N₂O using the conversion factor of 1.57111 kg N₂O/kg N₂O-N.

Indirect nitrous oxide emissions result from volatile nitrogen losses that are mineralized to ammonia nitrogen during manure management. These emissions were calculated using the EF of 0.197 kg NH₃ N/kg N_{TAN} for house and 0.15 kg NH₃-N/ kg N_{TAN} for emissions from slurry-based systems. They are based on national animal place-based emission factors from Döhler et al. (2002), which were converted to TAN reference by Dämmgen et al. (2010). Emissions of NH₃ from slurry-based systems are calculated based on the amounts of N from the barn after subtracting the amounts of N emitted there. For ammonia emissions during grazing, an EF of 0.1 kg NH₃-N/kg N_{TAN} (EMEP 2013) based on the amount of TAN excreted is applied. A 10% transformation between each fraction of the N_{org} - and TAN pools was assumed (Haenel et al. 2020). The total NH₃ amount is calculated by multiplying by the conversion factor of 1.21587 kg NH₃/kg NH₃-N related to the NH₃-N amount. The calculation of the indirect N2O amount is done by multiplication with the EF from IPCC (2006) of 0.01 kg N₂O-N/kg NH₃-N related to the NH₃-N amount.

Annual CO_2 emissions from electricity and process water were included. For all variants in the study, a standard value according to (KTBL 2016) of 70 kWh/cow for electricity consumption and 31 m³/cow for process water consumption was included. Energy production and emissions related to feed manufacturing are not included in the study.

Calculation procedure

The carbon footprint was calculated for three feeding strategies using the data from meta-analysis and EF described above in the REPRO model. In the second step, we investigated how the carbon footprint of milk was affected by varying individual parameters such as the level of enteric methane emissions, milk yield, and feed conversion. The enteric CH_4 emissions were decreased for -8.8%, the ECM yield was increased for +4.1% and feed efficiency was +4.4%.

Results

Total GHG emissions for milk were calculated to be for ration 1 (CON): 0.41 kg CO_{2eq} /kg ECM, for ration 2 (CON): 0.39 kg CO_{2eq} /kg ECM, and for ration 3 (CON): 0.56 kg CO_{2eq} /kg ECM (Fig. 3).





While there are almost no differences among rations 1 and 2 in their respective variants, ration 3 produces much higher emissions throughout all variants. Within all three rations, the total emissions decrease from variant 1 (CON) to variant 3 (-15% CH₄ reduction).

The findings confirmed that GHG emissions can be reduced by up to 6% solely through the reducing effect of EO on methane production. The results also showed that when taking more variable parameters into account such as the positive effects on milk yield and feed efficiency, a GHG reduction potential of up to 10% can be achieved. Due to the high contribution of enteric methane emissions to the CO_2 balance of milk, it has a significant impact on final results. Overall, enteric CH_4 emissions accounted for the largest share of CO_2 emissions per year (average of all variants: 64%). By reducing the contribution of enteric methane emissions to the total GHG emission, the relative contribution of enteric CH_4 emission results in 62%; meanwhile, CH_4 emissions from manure increase slightly from 24 to 25%.

The detailed results of sub-indicators per cow and year in milk production are summarized in Table 5. The results show a degree of differentiation between the three rations,

Table 5 Annual and product-related CO_{2eq} emissions per cow and year and per annual produced energy-corrected milk per year depending on ration type and associated variants

		Feed ration								
		Ration 1			Ration 2			Ration 3		
		CON ^a	EO ^b	-15% CH ₄ ^c	CON	EO	- 15% CH ₄	CON	EO	-15% CH ₄
Edible protein (eP)	kg eP/y	372.6	387.9	372.6	372.6	387.9	372.6	372.6	387.9	372.6
Milk yield	kg/y	10,084	10,549	10,084	10,084	10,549	10,084	10,084	10,549	10,084
ECM yield ^d	kg ECM/y	10,035	10,497	10,035	10,035	10,497	10,035	10,035	10,497	10,035
Emissions										
CH ₄ enteric fermentation	kg CO _{2eq} /y	2765	2522	2350	2615	2385	2223	3291	3001	2797
CH ₄ excreta	kg CO _{2eq} /y	1074	1074	1074	1099	1099	1099	985	985	985
N ₂ O direct	kg CO _{2eq} /y	-	-	-	-	-	-	1097	1097	1097
N ₂ O indirect ^e	kg CO _{2eq} /y	100	100	100	86	86	86	70	70	70
Energy use ^f	kg CO _{2eq} /y	144	144	144	144	144	144	144	144	144
GHG emissions	kg CO _{2eq} /y	4082	3839	3668	3944	3713	3551	5586	5297	5093
GHG emissions	kg CO _{2eq} /kg ECM	0.407	0.366	0.365	0.393	0.354	0.354	0.557	0.505	0.507

^aCON, control variant

^bEO, variant with essentioal oils blend

^cVariant with a reduction of 15% enteric methane emissions

^dAmount of energy-corrected milk per year

^eMainly from manure management

^fElectricity and process water used on farm

both in terms of overall impact and in terms of the contribution of each sub-indicator as emission source.

Carbon footprint contributions from different stages and gases differed between barn rations with grass and corn silage and pasture rations. When considering gas contribution, enteric CH_4 accounted for the largest contribution, while NH_3 had the lowest one.

The contribution of enteric methane to GHG emissions was greater in the barn than in the grazing ration. The CH_4 amounts are also calculated in similar proportions for the stable rations (ration 1 (CON): 68%, ration 2 (CON): 66%, ration 3 (CON): 59%).

 CH_4 emissions from manure management are in second place in the contribution to GHG emissions and are again very similar for the stable rations (ration 1 (CON): 1074 kg CH_4/y , ration 2 (CON): 1099 kg CH_4/y). For the pasture ration, on the other hand, a CH_4 emission potential of 985 kg per year was calculated.

 N_2O emissions from pasture were calculated at 1097 kg CO_2 , resulting in a contribution to total GHG emissions of 20% for this ration.

The ammonia-related CO_2 emissions from barn, manure storage, and pasture differ only slightly between ration types 1–3. Ration 1 achieves the highest results with 100 kg CO_{2eq} /year, due to the high N intake.

There is no difference in the results of energy intensity since the energy expenditure of the husbandry system, manure storage systems, and the direct use of energy is the same in all feed rations. The emissions from energy intensity were calculated to be 144 kg CO_{2ed} /y.

The total GHG emissions per year in this study decreased with increasing ration share of maize silage by a difference of 13.8 kg CO_{2eq} /t ECM.

Among the various noteworthy results are also the calculated CO_{2eq} emissions in relation to the amount of energycorrected milk produced. Figure 4 shows the product-related CO_{2eq} emissions (g CO_{2eq} /kg ECM) and the various sources that contribute to it in more detail.

Enteric methane emissions

Calculated methane emissions from enteric fermentation were for ration 1 (CON) 275.6 g CO_{2eq}/kg ECM. With increasing corn silage content, the results in ration 2 (CON) were lower at 260.6 g CO_{2eq}/kg ECM. Ration 3 (CON) achieves the highest value of enteric methane emissions (enteric stable: 268.6 g CO_{2eq}/kg ECM and enteric pasture: 59.3 g CO_{2eq}/kg ECM), which is due to the high crude fiber content of the alfalfa hay used in summer and winter feeding. Due to the alfalfa hay, the ration has a higher crude fiber content, which in turn increases the potential for enteric CH₄ emissions. Variant 2 of rations 1–3 each shows the reduction in total methane production due to an addition of EO, and the associated lower enteric methane output per day, amounting to – 8.8%.

More details of the calculated EF of methane emission from enteric fermentation are shown in Table 6.

Manure methane emissions

The results of GHG emissions from manure management are in a similar range, as a liquid system is used for all of them (ration 1 (CON): 107.0 g CO_{2eq} /kg ECM, ration 2 (CON): 102.3 g CO_{2eq} /kg ECM). Overall, ration 3 (CON) has the lowest GHG emissions of 97.0 g CO_{2eq} /kg ECM from manure management and 1.1 g CO_{2eq} /kg ECM from grazing.



Fig. 4 Product-related CO_{2eq} emissions from methane emissions (enteric fermentation, manure liquid storage, and pasture) depending on ration type and associated variants

Table 6Calculated emission factors of enteric methane emissions ofdifferent ration types (ration 1 grass-fed, ration 2 corn-fed, ration 3with pasture) normalized to 1 day, 1 year, 1 kg DMI, 1 kg ECM and1 kg VS

Emission factors of	Feed Ration						
enteric methane emission	Ration 1	Ration 2	Ration 3				
	CON ^a	CON	CON				
kg CH _{4-enteric} per year	111	105	132				
kg CH _{4-enteric} per kg DMI ^b	5.5	5.3	6.5				
g CH _{4-enteric} per kg ECM ^c	11.0	10.4	13.1				
g CH _{4-enteric} per kg VS ^d	27.9	28.8	22.3				
g CO _{2eq-enteric} per day	7576	7164	7383				
kg CO _{2eq-enteric} per kg year	2765	2615	2695				
kg CO _{2eq-enteric} per kg DMI	138	132	163				
g CO _{2eq-enteric} per kg ECM	275.6	260.6	268.6				

^aCON, control variant

^bDMI, dry matter intake

^cECM, energy-corrected milk

^dVS, volatile solids

Discussion

Life cycle assessment studies that compare the carbon footprint of milk and analyze the effects of changing input parameters are rare. Therefore, the calculated results are discussed with studies that analyze the contribution of parameters to the carbon footprint calculation in different milk production systems and feed rations.

A discussion of results of calculated carbon footprints of milk is difficult because different models and different assumptions are used (Rotz 2018). Many comparable models for greenhouse gas accounting calculate with highly simplified approaches to represent the complex physiological processes in the animal and the material transformation processes in the barn, manure storage, and pasture (Flysjö et al. 2011). Thus, the use of general EFs cannot adequately capture relevant uncertainties associated with natural variations in biological systems. In this study, specific EFs were calculated except for direct and indirect nitrous oxide emissions. For the EF calculation of enteric and storage methane emissions, ration-related components are considered, which are important input variables in addition to dry matter intake. The basis for methane emissions from manure management is the DMI, the crude ash content, and the amount of fermentable matter in the excrements. Direct and indirect nitrous oxide emissions are calculated as a function of the amount of nitrogen ingested and the type of husbandry system in place.

The level of GHG emissions from dairy production is reported in comparable studies between 0.6 and 1.8 kg

 CO_{2eq}/kg ECM (Gerber et al. (2011), Rotz and Thoma (2017), Famigliette et al. (2018)). Since only emissions from animal husbandry, manure storage, grazing, and direct energy from electricity and industrial water were included, the emission intensities of this study are lower, with values ranging from 0.4 to 0.6 kg CO_{2eq}/kg ECM. Feed production and milk production were not considered, as mentioned at the beginning.

Comparing the results of variants 1 and 2, it is noticeable that GHG emissions decrease with increasing ECM yield of variant 2. This correlation can be confirmed by the study of Rotz (2018). Another reduction factor is the better feed conversion in variant 2 of the investigated feed rations. It also represents an important key factor for reducing the product-related carbon footprint, as stated by O'Brien et al. (2014), among others.

Also, Rotz (2018) found that grazing ration had the highest total GHG emissions per year and the highest productrelated GHG per kg ECM in his study compared to yearround confinement. In his study of representative dairy farms in Ireland, he calculated a pasture-based emission factor of 1.21 kg CO_{2eq} /kg ECM. As in this study, he describes proportionally higher values of enteric methane and nitrous oxide emissions for grazing. When classifying the result, it should be noted that Rotz (2018) includes feed-related CO_2 emissions in his calculations. In this study, ration type 3 with pasture reaches a value of 0.557 kg CO_{2eq} /kg ECM and also the highest GHG emissions compared to the two rations without pasture.

The level of potential ammonia emissions is influenced by the amount of forage nitrogen ingested, because the higher the crude protein concentration of the ration, the higher the N excretion (Burgos et al. (2010), B. Frank et al. (2002)). In particular, the amount of ammoniacal nitrogen in urine is crucial in the calculation of stall- and store-related NH₃ emissions. Comparing the rations without pasture, it is noticeable that higher CO₂ emissions from ammonia were calculated for ration 1 because it has a higher crude protein content. Similar results are also shown by B. Frank and Swensson (2002) in their study.

The calculation of enteric methane emissions is fundamental to the carbon footprint of milk, as they account for the largest share of GHG emissions from dairy production O'Brien et al. (2014). It describes results depending on the orientation of the feed ranging from 59% for pasture to 68% for barn feeding.

There are different models for estimating enteric methane emissions based on either dry matter intake, raw nutrients, milk yield, and live weight (Rauen 2018). In this study, enteric methane emissions were estimated using an emission factor calculation based on raw nutrient contents and store-specific parameters. With respect to enteric methane emissions, variations in ration-related input factors lead to significant changes in the emission pattern, allowing the identification of reduction potentials through different ration strategies. Rations 1 (grass-based) and 2 (corn-based) with have slightly lower GHG emissions from enteric fermentation than ration 3 with pasture with 300.8 g CO_{2eq}/kg ECM and 288.8 g CO_{2eg}/kg ECM, respectively. Thus, the assumption that the high crude protein intake on pasture leads to a higher CH₄ production compared to rations 1 and 2 could be confirmed. Comparing the two rations without pasture with each other, it can be seen that the higher crude protein content in ration 1 has a negative effect on enteric methane emissions. Hülsbergen and Rahman (2013) calculated a GHG potential from enteric CH₄ emissions of 326 g CO2eg/kg ECM for conventional and 419 g CO2eg/kg ECM for organic dairy farms with pasture. In contrast to our study, the calculations of Hülsbergen and Rahman (2013) were based on the formula of Ellis et al. (2007) in which dry matter intake is integrated. Comparisons of the results of this study with those of Hülsbergen and Rahmann (2011), Flysjö et al. (2011), Rotz (2018), and the standard IPCC (2006a, b) emission factor nevertheless suggest that the formula is sufficiently accurate.

Comparing the results of storage-related methane emissions of rations 1 and 2, it is evident that increasing the amount of corn in the feed ration is a strategy to reduce CH_4 emissions (Garnsworthy et al. (2012), Wilkinson and Garnsworthy (2016), Hristov et al. (2013), Hassanat et al. (2013)). Comparable results were shown by Van Middelaar et al. (2013) and Wilkinson and Garnsworthy (2016). The use of whole-crop corn silage increases forage digestibility due to its higher starch content and thus contributes significantly to the reduction of storage-related CH_4 emissions.

Across all manure systems, most studies of potential emissions focus on CH_4 emissions, as it has the largest contribution as a climate gas to the CF of milk (Cárdenas et al. 2021). Our results are similar to data in the literature; for example, Misselbrook et al. (2016) found a value of 34.3 g CH_4 /kg VS for annual emission, while Petersen et al. (2016) found daily values of 31.2 g CH_4 /kg VS and 11 g CH_4 /kg VS from slurry pits with residence times of 30 days in an in vitro study.

Regarding the storage-related methane emissions, the digestibility of organic matter and the crude ash content in the ration are of central importance. Expressed in CO_2 equivalents, the highest values are observed in ration type 1 with 1074 kg CO_{2eq} /year, while ration type 2 reaches a similar level with 1099 kg CO_{2eq} /year. The comparatively lower value of ration type 3 of 985 kg CO_{2eq} /year results from the favorable ratio of VQ OM, the lower crude ash values, and the lower MCF value of 0.01 of pasture compared to manure-based systems.

It is well documented that mitigation strategies, to reduce enteric CH_4 emissions, can have a large impact on total GHG emissions (Beauchemin et al. (2009), Benchaar and Greathead (2011), Castro-Montoya et al. (2015), Ugbogu et al. (2019)). Various production practices, such as increasing feed digestibility and digestible feed intake or reducing enteric methane production by adding free oils or oil-rich feeds, biologically active plant compounds such as EO, tannins, and saponins, or improvements in feed quality, aim to reduce methane emissions.

In this study, the focus was on the CH_4 reduction potential of essential oils and its effect on the GHG balance of milk. The results of this study confirmed that GHG emissions can be reduced by up to 6% per capita per day due to the reducing effect of EO on methane production. Considering further variable parameters, such as the positive effects of EO on ECM yield and feed efficiency, a GHG reduction potential of up to 10% can be achieved for the grass and corn silage ration and almost 9% for the grazing ration. Only a small number of in vivo experiments have also described CH_4 mitigation potential of essential oils (Tager and Krause (2011), Castro-Montoya et al. (2015), Cobellis et al. (2015), Klop et al. (2017), Elcoso et al. (2019), Hart et al. (2019)).

Conclusion

The main objective of the present work was to analyze the impact of changing different factors in the carbon footprint calculation of milk. Considering the results of a meta-analysis by Belanche et al. (2020a, b), enteric methane emissions per day (-8.8%), ECM performance (+4.1%), and feed efficiency were changed by +4.4% using the essential oil blend Agolin Ruminant. The results of the current study showed that there is a positive effect of Agolin Ruminant on the carbon footprint of milk (kg CO_{2eq}/year) in relation to the amount of ECM produced (kg CO_{2eq}/kg ECM) of 10%.

Author contribution All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Franziska Becker, Katrin Spengler, Clara Heider-van Diepen, and Frank Reinicke. The first draft of the manuscript was written by Franziska Becker, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Data availability All data and materials, as well as software applications, support the published claims. Datasets related to this article can be found at Belanche, A., Bach, A., Morgavi, D. P., Newbold, C. J., Yánez-Ruiz, D. R., and Zweifel, B. (2020) A Meta-analysis Describing the Effects of the Essential oils Blend Agolin Ruminant on Performance, Rumen Fermentation and Methane Emissions in dairy Cows. Animals 10, 620. https://doi.org/10.3390/ani10040620.

Declarations

Ethics approval Ethical approval is not applicable for this article.

Consent to participate In this study, no participants were involved.

Consent to publish All authors have approved the manuscript before submission and agree to publication.

Competing interest The authors declare no competing interests.

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