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Cradle-to-grave life cycle assessment of buffalo mozzarella cheese supply chain *in central Italy*

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ABSTRACT

Public opinion about environmental aspects is generating a widespread concern, which contributes to lead consumers to choose sustainable products. Livestock systems are responsible for the emissions of different pollutants and, as a consequence, this is reflected in the environmental sustainability of animal-based products.

This study was aimed at evaluating the environmental impacts of buffalo mozzarella cheese with a cradle-to-grave life cycle assessment approach from the primary production of milk to the end-of-life of buffalo mozzarella cheese. The functional units were 1 kg of buffalo Fat and Protein Corrected Milk (FPCM) evaluated at farm gate and 1 kg of consumed buffalo mozzarella cheese evaluated at consumers' home.

The study enrolled 9 buffalo dairy farms, 2 dairy plants and 4 markets where 243 customers were surveyed. Processes of raw milk production, mozzarella cheese making, transports, retail and home consumption were considered. The accounted environmental impacts were: climate change (CC), terrestrial acidification (TA), freshwater eutrophication (FE), marine eutrophication (ME), ozone depletion (OD), ecotoxicity (EC), human toxicity (HT), land use (LU) and fossil depletion (FD). Buffalo farming was the primary hotspot accounting for more than 50% throughout all impact categories and having a major role in CC, TA, FE, ME, OD and LU. The energy consumption of cheese making had a contribution of 29% in HT, whereas the use of high-emitting vehicles by consumers accounted for 23% and 17% in EC and FD, respectively. To reduce several sources of pollution and mitigate the environmental impacts of buffalo mozzarella cheese, the LCA hotspots identification can lead to the development of specific mitigating strategies to improve sustainability along the supply chain. The development of farm-based models may better assess environmental impacts, the mitigation achieved by the implemented best practices and the "pollution swapping" effect to evaluate the effectiveness of the mitigation strategy.

1. Introduction

Nowadays consumers have strengthened their attention on agriculture and livestock's environmental impact and the relatives' effects. This common awareness leads to the need of additional characteristics to define sustainable products.

Livestock systems contribute at releasing of several pollutants by affecting different compounds such as water and air quality, and depleting natural resources such as water and soil [1].

Among the environmental impacts associated with livestock systems, the contribution of greenhouse gases (GHG) to climate change has been the most discussed in the last years.

The Sixth Assessment Report [2] indicates livestock, among anthropogenic activities, as a large source of methane emission mainly referring to enteric fermentation that depends on different factors such as the type, amount and quality of feed, the amount of meat and milk production and the animal health and growth rate. In addition, manure management is a relevant source of CH_4 and nitrous oxide (N_2O) emissions.

Globally, enteric fermentation and manure management are responsible for 48% and 7% of the total GHG emissions of the agriculture sector, and the larger source of these emissions refers to dairy and

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non-dairy cattle, followed by buffaloes, sheep and goats [3].

In Italy, 7.8% of GHG emissions are attributable to the agriculture sector. The CH_4 from enteric fermentation is the primary source of GHG with 45% of the contribution, followed by emissions from managed soils and manure management that sharing 28% and 20%, respectively [4].

Besides GHG emissions, agriculture is the largest source of ammonia (NH_3) accounting for 75% of the global emissions and contributing to the aerosol burden [2].

Dairy supply chain is employed in milk and cheese production, additionally to meat as a co-product, and is gaining more attention for improving the environmental performance of the dairy industry.

Buffalo farming accounts for 15% of the global milk yield (2021) and it is the second contributor after cows' milk to the global dairy market. This species is gaining more attention due to its strategic role in the economy and society. Buffalos' features allow to have higher efficiency to convert fiber into energy and to best adapt in hot-humid areas critical for other ruminants [5,6].

The 87% of the European buffalo population is reared in Italy in intensive systems, followed by Romania and Bulgaria with a contribution of 3% [7].

The buffaloes belonging to the Mediterranean Italian Buffalo breed reached a total of 435,134 heads in 2023, mainly farmed in the Centre and South of Italy. The Italian buffalo farming is increasing over the years and this positive trend shows a +7.6% of reared buffaloes from 2018 to 2023 [8].

Buffalo milk has a different composition compared to cows and sheep milk, presenting a richer content of fat and protein, and it is employed in many Italian typical products [9]. The characteristic composition of buffalo milk and the traditional manufacturing practices of a defined geographical area in Italy allowed for the declaration of Protected Designation of Origin (PDO) for "Mozzarella di Bufala Campana". Italian Buffalo mozzarella cheese reached 55,815 tons in 2022 and represents the product whose production showed a continuous increase over the years recording a +13% from 2018 to 2022 [9].

Life Cycle Assessment (LCA) is a common method used to evaluate the potential environmental impacts connected to the life cycle of a product or service and is standardized by ISO 14040 and ISO 14044 [10, 11].

Few LCA studies assessed the environmental consequences of buffalo dairy production. Pirlo et al. [12] investigated the environmental impact of buffalo milk from six dairy farms in Southern Italy and reported that the most emissive activities were enteric fermentation, manure management and purchased feed production at farm gate. Chirone et al. [13] showed that the main drivers for most of the environmental impacts of buffalo milk at farm gate were the purchased feed and machine operations, whereas enteric methane and ammonia emission were relevant to climate change, terrestrial acidification and terrestrial eutrophication. In addition, these Authors emphasized the role of the allocation method in the final result indicating that the economic allocation gives a larger share of environmental impacts to milk production compared to a biophysical approach.

Other authors [14] studied the role of managing growing animals in the environmental assessment of buffalo milk at farm gate and reported that the free-ranging system of heifers reduces water consumption and other pollutants compared to the confined ones.

Garg et al. [15] evaluated the carbon footprint of milk production in a multi-functional smallholder dairy buffalo system in western India, and pointed out the relevant contribution of enteric methane for the small systems.

Soares et al. [16] investigated the effect of different feeding scenarios on the environmental impact of buffalo milk production in Brazil and, in this context, the extensive buffalo breeding scenario was the most impactful for climate change, land use and water consumption.

The LCA case study of Italian buffalo mozzarella cheese was assessed by Berlese et al. [17] who considered the life cycle from cradle-to-plant-gate and highlighted the contribution of buffalo milk production under different scenarios for the impact categories of global warming, acidification and eutrophication.

To date few studies assessed the LCA of buffalo milk and only one study assessed the LCA of buffalo mozzarella cheese at plant gate but no studies evaluated the whole supply chain of buffalo mozzarella cheese up to final consumption. Therefore, there is a lack of knowledge of the retail and consumption stages of the buffalo mozzarella cheese supply chain. When these stages were analyzed for the bovine mozzarella cheese supply chain [18,19] it was pointed out that energy consumption was the most contributor to environmental impacts.

In light of what stated above, this study was aimed to improve the knowledge on the environmental performance of buffalo mozzarella cheese supply chain referred to the Centre of Italy throughout the whole cycle, from raw milk production to home consumption.

2. Materials and methods

2.1. Goal and scope

The main goal of this study was to estimate the environmental impacts of buffalo mozzarella cheese by evaluating raw milk production, dairy plant operations, distribution, retails and consumer habits. The dairy supply chain was analyzed by following the ISO 14040/14044 methodology and considering the product with a "from cradle to grave" LCA approach. The final scope of this study was the analysis of hotspots to identify the most impactive sources throughout the buffalo mozzarella cheese produced and supplied in the context of central Italy and to provide insights of mitigation practices.

2.2. Functional units

The functional units were 1 kg of buffalo fat and protein corrected milk (FPCM) evaluated at farm gate and 1 kg of packaged buffalo mozzarella cheese with a moisture content of 61% evaluated after home consumption.

To transform raw milk into FPCM the following equation was used [20]:

$$FPCM\ (kg/y) = (\{[(g\ of\ fat/L - 83.0) + (g\ of\ protein/L - 47.3)] \times 0.00687\} + 1) \times milk\ yield\ (kg/y) \quad (1)$$

The reference milk fat and protein content adopted in the present study were 8.30% and 4.73%, respectively.

2.3. System boundaries

The system boundaries included raw milk production, cheese making, distribution, retail and home consumption. The farm activities for raw milk production considered the feed production (on and off-farm), animal management and the transports of auxiliary products (mineral fertilizers, extra-farm feeds). Raw milk transports, cheese making, packaging and the distribution of buffalo mozzarella cheese to the retails were activities within the dairy plant. The selling and consumption encompassed the habits of purchasing, transports to home and home packaging disposal. All the stages of buffalo mozzarella cheese production and consumption were located in the Centre of Italy. The system boundaries of the mozzarella LCA study and the related unit processes are represented in Fig. 1.

2.4. Significance analysis

Once system boundaries were defined, the first step was to identify the key processes, parameters, and elementary flows that substantially contributed to the environmental impacts. This study aimed to minimize cut-offs wherever possible. Nevertheless, cut-offs were permissible under certain conditions that involved comprehensive evaluation of

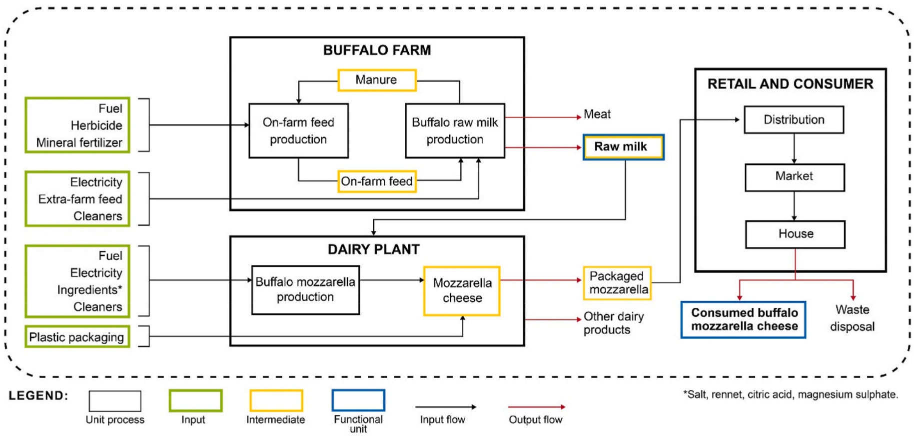

Fig. 1. System boundaries of buffalo mozzarella cheese supplied chain.

mass, energy, environmental significance, and cumulative effects [10, 11]. Adhering to these guidelines, a significance analysis was conducted to identify the most relevant issues and selectively exclude processes with minimal influence on the overall conclusions or the intended application of the study.

While every process was typified by material requirements and energy flows, elements such as farm buildings, machinery, veterinary accounting, and employee commuting were excluded from the assessment. Electricity consumption at markets was not considered, given that mozzarella conservation did not occur in cold storage. Energy and refrigerant systems leakages from home and energy for cooking were excluded from the system boundaries. Buffalo mozzarella cheese is consumed fresh, for this reason does not need cold storage and cooking operations.

In this study, the changes in soil organic carbon (SOC) stocks were not accounted for, based on the widely accepted scientific tier 1 premise that SOC, under specific climate and management conditions, tend towards an equilibrium [21].

The result was an inclusion or elimination of information from the case study. Extended data can be found in the supplementary materials (Table S1).

2.5. Life cycle inventory

The study enrolled 10 commercial dairy buffalo farms, 2 cheese making plants, 4 markets and 243 consumers. Data were collected throughout the year 2022.

Each participating part was surveyed with specific questionnaires regarding farm management (crop production, herd composition, animal feeding, milk production, housing system, and manure management), cheese making (processed raw milk, energy consumed, cheese yield, and packaging information) and consumer habits (type and amount of cheese purchased, type of transport, distance travelled and waste disposal). When primary data were not available, secondary data were taken from Ecoinvent v3.2 database [22] and literature [21,23].

During the data collection, one commercial dairy buffalo farm was increasing the number of animals and declared heifers were not compliant with culling rate. We considered this condition not representative and the farm was excluded from the study. Therefore, a total of nine farms were evaluated.

To ensure the validity of primary data, the Ecoinvent pedigree matrix approach was adopted [24] by assigning specific distributions of uncertainty at the input dataset through flow indicators addressing source reliability, temporal, geographical, and technological correlation, and data sampling methods. These flow level indicators addressed the level of review that the unit process had undergone and the completeness of the unit process. Results of the pedigree matrix can be consulted in the supplementary materials (Table S2).

2.5.1. Buffalo farms

The nine dairy farms belonged to the productive district of Amaseno (FR) located about 80 km south of Rome. This area represents an important Italian site for buffalo farming with a total of 23,043 heads [8] which represents about 5.5% on the national scale.

In terms of herd size, the investigated farms had 272 $\pm$ 127 heads on average and they fall into the class of 100–500 heads that represents 59% of Italian dairy buffalo farms and 57% of the national dairy buffalo population as reported in the National Livestock Database [8].

The buffalo district of Amaseno is a suitable area for a PDO mozzarella label [25]. However, not all buffalo farmers in this area join the PDO consortium, and this is the case of buffalo farms and cheese making plants enrolled in the present study.

Although the selected farms had differences in herd size, they were homogeneous in terms of crops cultivated, housing system, animal genotype and farm management. Buffaloes were fed with Total Mixed Ration (TMR) twice a day after milking operations and the TMR was mainly based on self-produced cereal silage (corn, triticale and sorghum) and hay, whereas concentrate feed was purchased and principally used for lactating adults and fattening calves. Additional information on diets is reported in Table S3 of supplementary materials.

Six farms used to treat the manure before field spreading, whereas the other three farms stored untreated manure without any additional operations. The manure treatment occurred with a slurry separator where solids were mechanically separated from the slurry. The liquid fraction was stored in a slurry tank and the solid part in a pile. One of the six farms combined slurry separation with the anaerobic digestion of the liquid fraction. The digested slurry was used for organic fertilization together with the separated solid part.

Table 1 summarizes the key inventory information of the 9 investigated farms.

Farmers had an average cultivated area of 33 ha characterized by at least an annual crop rotation of autumn-winter and spring-summer cultivation to support most of the animal's nutritional needs. The soil operations had high variability between the farms and ranged from a minimum tillage with less than 10 cm depth and an intensive tillage having 40 cm depth.

Manure was mainly applied through surface spreading to all crops, although some farms used to incorporate it in depth. Inorganic fertilization such as ammonium nitrate, urea and ammonia phosphate were provided mostly for sorghum and corn.

Electricity consumption was referred to the milking parlor, cooling system (fans) and milk storage, whereas diesel usage was mainly due to feeding distribution, soil tillage, crop harvesting and irrigation operations.

Table 1
Main data of the nine commercial dairy buffalo farms enrolled in the study.

		Farms									
		1	2	3	4	5	6	7	8	9	**Mean±SD**
Herd	Dairy buffalo, no	178	69	118	149	102	302	152	152	233	**162 ± 70**
	Dry buffalo, no	46	18	37	39	35	79	52	40	61	**45 ± 17**
	Heifers, no	67	26	35	45	31	113	46	68	70	**56 ± 27**
	Beef calves, no	12	–	–	–	–	50	–	25	–	**10 ± 17**
	Milk, L/day/head	9.33	8.02	8.27	8.78	8.71	8.91	8.24	8.23	7.69	**8.46 ± 0.51**
	Fat, %	8.17	8.45	7.79	7.83	8.37	8.02	8.13	7.96	7.55	**8.03 ± 0.29**
	Protein, %	4.73	4.83	4.69	4.51	4.86	4.70	4.78	4.72	4.62	**4.72 ± 0.11**
	Forage in diet as DM, %	65	81	64	65	58	78	71	76	69	**70 ± 7**
Crop	Cultivated area, ha	26	14	30	25	15	73	45	37	29	**33 ± 18**
	Cereal silage, ha	18	14	20	7	3	47	20	23	20	**19 ± 12**
	Hay, ha	8	–	10	18	12	26	25	14	9	**15 ± 7**
	Mineral fertilizer, kg N/ha	70	95	60	45	65	95	60	95	130	**79 ± 26**
	Mineral fertilizer, kg P/ha	–	60	30	–	–	–	40	–	40	**19 ± 24**
	Organic fertilizer, kg N/ha	210	245	240	255	235	205	240	205	150	**221 ± 32**
	On-farm feeds, t/y	1674	787	1353	595	285	3992	1725	2040	1730	**1576 ± 1085**
	Extra-farm feeds, t/y	696	115	482	1236	1219	1345	427	684	1329	**837 ± 456**
Energy	Electricity, MWh/y	48	19	32	40	28	81	31	41	57	**42 ± 19**
	Diesel, t/y	31	16	21	18	15	79	20	40	42	**32 ± 21**

Data on herbicides, milking parlor cleaning solutions and refrigerant systems are consultable in the supplementary materials (Table S4).

2.5.2. Dairy plants

Two cheese making plants processing buffalo milk from a sample of investigated farms were enrolled in the study. The dairies differ in the size, the bigger one was industrial based dairy plant that processed a large amount of milk produced from some of the investigated buffalo farms and here indicated as n° 1; the smaller one was a farm-based cheese making plant that processed only the milk produced in the same farm and here indicate as n° 2. Both dairies can be considered representative of the buffalo mozzarella produced in the area in term of cheese making processes and mozzarella cheese yield (28%). Main data of dairy plants are shown in Table 2.

Cheese making plants produced mozzarella cheese in various sizes (250 g, 500 g, 1 kg, 2 kg) and packed in plastic bags of different weights. A representative packaging was calculated as the average of the weights of single bags needed to pack one kg of buffalo mozzarella cheese. Then, the total mass of packing was calculated by multiplying the representative bag to the total annual mass of mozzarella cheese produced.

Table 2
Main data of the two dairy plants enrolled in the study.

	Dairy plant		
	1	2	Mean
Production			
Buffalo raw milk, t/y	984.0	229.5	**606.7**
Processed milk for mozzarella, t/y	925.0	204.8	**564.9**
Processed milk for other cheese, t/y	59.0	24.7	**41.9**
Mozzarella cheese, t/y	259.0	57.3	**158.2**
Auxiliary materials[a] t/y	34.4[b]	8.0	**21.2**
Refrigerant cell, m^3	24	10	**17**
Packaging			
HDPE, t/y	1.63	0.49	**1.06**
HDPE, g/kg mozzarella	6.2	8.6	**7.4**
Energy			
Electricity, kWh/y	109,091	89,851	**99,471**
Diesel t/y	20.4	3.4	**11.9**
Transports			
Auxiliary materials, km/single-trip	115	71	**93**

[a] This section included ingredients (citric acid and salt) and cleaners.
[b] Primary data were not available. Values were esteemed starting from the information acquired by the interview of the dairy plant n°2.

Energy consumption at dairy plants referred to diesel and electricity requests for machinery operations and refrigeration systems associated with the whole dairy production.

Although dairy n° 1 processed much more milk compared to dairy n° 2, the difference in annual electricity consumption was not so relevant. Considering that the industrial dairy had more efficient manufacturing equipment and diesel was used for several machinery, the electricity consumption results lower than the farm-based dairy plant.

This stage included the transports of mozzarella cheese from the plant-gate to retails. The distribution network covered three regions of central Italy throughout several markets located between 25 and 250 km from the dairy plants. These transports were characterized by the distance between the dairies and the marketplaces, and the related amount of transported mozzarella. The baseline vehicles used for delivery were refrigerated lorries.

2.5.3. Consumers stage

Four cheese markets were selected to analyze consumer habits. Two of them were the point of sale of dairy plants enrolled in the study (indicated as "a" and "b"), and the other two were typical city supermarkets (indicated as "c" and "d") supplied by the selected dairies and located in a big city and in a midsized city to guarantee the representativeness of consumer sample and to better characterize the supply chain. A total of 243 consumers were surveyed about their purchasing and consumption habits; the extended questionnaire is consultable in supplementary materials (Section S5). Information on the amount of buffalo mozzarella cheese purchased, total shopping cost, type of transport, distance travelled from home to market, buffalo mozzarella cheese wasted, and packaging disposal were acquired. According to the most used customer practice, packaging was disposed as plastic waste sorting.

Table 3 reports the average habits for the investigated retails.

2.6. Allocation criteria

Buffalo farms are multifunctional systems that coproduced raw milk and meat from culled females and fattening animals. To ascribe the impacts between these two products was applied a biophysical allocation method defined by the International Dairy Federation [26] for dairy systems. The allocation factor was calculated as:

$$AF_{milk} = 1 - 6.04 \times BMR \quad (2)$$

where AF_{milk} is the allocation factor for milk and BMR is the ratio between the mass of live weight of all animals sold per year and the mass of

Table 3
Main data on purchasing habits provided by the customers interviewed.

	Customers habits				
	Customers interviewed at market				Total customers interviewed
	a	b	c	d	
Purchasing					
N° customers	50	9	84	100	243
Purchased mozzarella, kg	0.9 ± 0.6	1.7 ± 3.1	0.4 ± 0.3	0.5 ± 0.4	0.6 ± 0.8
Final shopping cost, €[a]	22.4 ± 12	29.4 ± 46	44.4 ± 28	52.5 ± 32	43 ± 30
Purchased mozzarella, %[b]	56 ± 22	69 ± 13	24 ± 23	26 ± 24	33 ± 27
Transport					
Distance travelled[c], km	15 ± 10.5	6.4 ± 8.5	4.1 ± 4.7	6.6 ± 6.7	7.4 ± 8.2
Transport type[d] (km)					
On foot	–	–	0.2 ± 0.03	0.2 ± 0.03	0.1 ± 0.02
Bicycle	–	–	–	0.03	0.012
Scooter	0.5 ± 0.5	–	–	0.3 ± 0.2	0.2 ± 0.3
Hybrid/electric car	1.04 ± 0	–	0.4 ± 0.3	0.8 ± 1.2	0.7 ± 0.9
Petrol car	3.1 ± 2.2	0.7 ± 0	1.4 ± 2.7	3 ± 3.5	2.4 ± 2.7
Diesel car	8.2 ± 5.6	5.4 ± 8.5	1.4 ± 1.2	1.3 ± 1.1	2.9 ± 3.4
Gas car	2.1 ± 1.5	0.3	0.7 ± 0.6	0.5 ± 0.3	0.9 ± 0.9
Bus	–	–	–	0.1 ± 0	0.02 ± 0
Metro/train	–	–	–	0.4 ± 0.4	0.2 ± 0.2

[a] Total expense at market.
[b] Percentage of mozzarella cost on total market expense.
[c] Average value calculated for each market as stated by consumers.
[d] This section referred to the contribution of each type of transport to the distance travelled for the market-home journey.

milk (as FPCM) sold per year. The correction of milk was previously assessed through the equation proposed by Di Palo [20] which considers the fat and protein content of buffalo milk.

The annual amount of live weight sold included the reformed female buffaloes, male calves for all the farms and, if available, fattening beef (up to 18 months).

Between the nine farms, this biophysical approach allocated the 78% of flows (e.g., gross energy intake, manure produced, organic and inorganic fertilisers, energy, transports, auxiliary materials) to raw milk (as FPCM).

The mass was considered to allocate flows (energy, cleaners, sanitisers, rennet, salt, citric acid, packaging, refrigerant gasses leak, other auxiliary materials) at cheese making plants between buffalo mozzarella cheese and other dairy products (ricotta cheese, yogurt, etc.). Considering the mass of all types of cheese produced, the 92% of flows were allocated to buffalo mozzarella cheese.

The flows of energy coming from the market-to-home transports were economically allocated considering the incidence of the expense for the buffalo mozzarella cheese (33%) on the total market expenditure.

2.7. Life cycle impact assessment

The method to calculate the GHG emissions at farm gate followed the approach proposed by IPCC [21] for cattle and buffalo. Enteric CH_4 emissions were calculated for each category of animal (lactating buffaloes and growing animals) using tier 2/3 methodology [21]. This is based on daily gross energy intake, digestible energy and the Y_m conversion factor corresponding to the fraction of gross energy converted to CH_4. Daily gross energy was calculated considering live weight, milk yield, activity, live weight gain and pregnant rate (%), while digestible energy was assumed as 55% for lactating buffaloes and fattening calves, and as 50% for dry buffaloes and heifers.

The Ym was estimated with a linear equation based on the amount of forage in the diet, which is a rough information of diet quality easily available. The linear equation was set starting from the information provided by IPCC [21] about the adoption of the Ym in relation to forage content in diet (Table 10.12). IPCC suggests to consider Ym of 7% for a diet with forage higher than 75%; Ym of 6,3% for a diet with forage comprised between 15% and 75%; Ym of 4% for a diet with forage comprised between 0 and 15%. Arbitrarily, we decided to consider a Ym of 7% for a diet with 80% of forage; Ym of 6,3% for diet with a 45% of forage and Ym of 4% for a diet with a 10% of forage.

The linear equation ($R^2 = 0.91$) was calculated starting from these three points as:

$$Ym = 0.042857143 * (x) + 3.8380952 \quad (3)$$

Where x is the amount of forage in the diet as % on dry matter. The estimated values of Ym for animal categories are reported in Table S3 of supplementary materials.

Solid and liquid manure, volatile solids (VS) and N yield yr^{-1} were calculated considering animal categories (lactating and dry buffaloes, heifers, calves, and fattening beef) and their housing system as reported in the Italian guidelines for manure management [27]. Country-specific conversion factors of 4.8 g and 15.3 g of $CH_{4\ per}$ kg^{-1} of VS and 0.02 and 0.05 kg of N_2O-N per kg N^{-1} for solid and liquid manure were used [28] to calculate CH_4 and N_2O (nitrous oxide) from manure storage, respectively. More details of the method can be consulted in our previous research [29].

At farm level were also accounted ammonia emissions produced by manure from housing and storage. According to CRPA [30], the 16% of total nitrogen excreted by animals in the barn was volatilized as NH_3-N, which was converted in NH_3 considering the ammonia molecular weight. Specific factors reported in the Italian Emission Inventory 1990–2018 [31] were adopted to calculate NH_3 emissions from manure storage. The emission factors were 23% and 14% of net nitrogen volatized as NH_3 from liquid and solid manure, respectively.

The emissions from crops were evaluated by using the DeNitrification DeComposition (DNDC) model [23] that was parametrized by local weather data, soil physical and chemical properties, organic and inorganic fertilisers, soil tillage and specific crops yield [32]. With this model was possible to account the nitrates (NO_3) and phosphate (PO_4^{3-}) lost by leaching and run off, the fraction of N volatilized as NH_3 and the emissions of nitrous oxide (N_2O). The list of the main DNDC emissions input/output parameters can be consulted in the supplementary materials (Table S6).

The emissions referred to the production and use of energy (electricity and fuel), extra-farm feed, inorganic fertilizers, herbicides, cleaning solutions, packaging and all transports occurred along the supplied chain were processed through the ReCiPe midpoint (H) method within the SimaPro v.9.1.1 software (Prè Consultants) by using the Ecoinvent database [22]. The relative emission factors are reported in Table S7 of supplementary materials.

To consider the annual leakage of refrigerant gas due to on-farm milk storage and dairy refrigerated cells was applied the equation defined by the Environmental Protection Agency [33] combining the refrigerant capacity and an annual leak rate of 10%.

Emissions related to packaging disposal were based on consumer habits, which were reported by the surveys. The most used practice was material recycling, that allow to re-use the plastic of the packaging to assemble new materials.

Since the cut-off approach's philosophy developed by Frischknecht [34] attributes the primary production's environmental burdens to the primary material user and excludes the recycled materials' potential credits from this consideration, in this study was just considered the transport of waste material from consumer home to waste plastic collector centre. A 50 km transport by truck was assumed to dispose mozzarella wasted packaging as reported by Dalla Riva et al. (2017). The environmental impacts related to the packaging end-of-life were assigned to the entities using the recycled plastic.

2.7.1. Impact of mitigation practices

Implemented practices that may mitigate GHG and ammonia were accounted with a farm-specific approach following the on-site characteristics. Table 4 gives a summary of the applied practices from the enrolled buffalo their potential emissions abatement.

2.7.2. Impact categories

To evaluate the environmental burden of buffalo mozzarella cheese supply chain were considered the following impact categories: climate change (CC; kg CO_2 eq.), terrestrial acidification (TA; kg SO_2 eq.), freshwater eutrophication (FE; kg P eq.), marine eutrophication (ME; kg N eq.), ozone depletion (OD; kg CFC11 eq.), ecotoxicity and human toxicity which include respectively terrestrial, freshwater and marine ecotoxicity, and human carcinogenic and non-carcinogenic toxicity (EC, HT; kg 1.4-DCB eq.), land use (LU; m^2a crop eq.), and fossil depletion (FD; kg oil eq.). All emissions were modelled with SimaPro v.9.1.1 software (Prè Consultants) by using the ReCiPe Midpoint (H) version 1.03 [35] and environmental impacts calculated [19]. The GHG emissions were converted to equivalent units of CO_2 according to the Global Warming Potential (GWP) in a 100-year time horizon. In the Recipe 2016 method, the GWP conversion factors per kg of CO_2, CH_4 and N_2O are 1, 34 and 298 kg, respectively.

In Table S8 of supplementary materials are reported the primary pollutants for the impact categories considered.

3. Results

The environmental impacts of 1 kg of buffalo FPCM at farm gate are reported in Table 5. The emissions related to the climate change (CC) were 3.03 ± 0.38 kg CO_2 eq., and the main sources were methane from enteric fermentation and manure management, nitrous oxide from manure management and feed production (on and off-farm feed), whereas energy and fuel usage provided a lower contribution. Terrestrial acidification (TA) accounted for 27.18 ± 4.44 g SO_2 eq. and the main source was ammonia released from manure management and crop fertilization. Freshwater eutrophication (FE) was 0.12 ± 0.03 g P eq. and it was mainly related to the production of off-farm feed. Marine eutrophication (ME) and ozone depletion (OD) were related to feed production, mainly on farm. Ecotoxicity (EC) and human toxicity (HT) were characterised by pollutants released by fuel combustion and herbicides. Land use (LU) and fossil depletion (FD) were associated with soymeal production and energy usage (electricity and fuel).

Table 4
Practices considered to evaluate the mitigation impact on GHG and Ammonia in the nine dairy buffalo farms enrolled in the study.

Mitigation practices	Farms								
	1	2	3	4	5	6	7	8	9
Manure management									
Slurry Separator[a]	x		x			x	x	x	x
Anaerobic Digestor[b]							x		
N field applications									
Injection at 6 cm depth[c]	x	x	x	x					
Feeding strategies									
Lipids supplementation[d]				x			x		x

The follow emission and abatement factors were considered in relation to management practices.

[a] -42% CH_4, +10% N_2O and +5%NH_3 [30,40].

[b] -67% CH_4, +41%N_2O and +31NH_3 [30,40].

[c] -75%, −49% and −95% NH_3 for nitrate and phosphate, urea and organic fertilizer [42].

[d] Each 1% of added lipids on diet (% of DMI) reduce 5.6% of enteric CH_4 [39].

Table 5
Environmental impacts expressed per 1 kg of FPCM (Fat Protein Corrected milk) evaluated at farm gate (**Mean±SD)** and per 1 kg of consumed buffalo mozzarella cheese evaluated at home.

Impact categories	Unit	FPCM at farm gate	Consumed Mozzarella cheese
CC	kg CO_2 eq	3.03 ± 0.38	12.03 ± 1.36
TA	g SO_2 eq	27.18 ± 4.44	102.20 ± 15.83
FE	g P eq	0.12 ± 0.03	0.62 ± 0.12
ME	g N eq	2.63 ± 0.95	9.45 ± 3.40
OD	g CFC11 eq	0.02 ± 0.002	0.06 ± 0.01
EC	g 1.4-DCB eq	1436.72 ± 451.43	8172.37 ± 1611.59
HT	g 1.4-DCB eq.	875.62 ± 261.97	4639.76 ± 935.23
LU	m^2a crop eq.	0.90 ± 0.34	3.25 ± 1.21
FD	g oil eq.	147.05 ± 17.91	882.61 ± 63.95

CC – Climate change, TA – Terrestrial acidification, FE – Freshwater eutrophication, ME – Marine eutrophication, OD – Ozone depletion, EC – Ecotoxicity, HT – Human toxicity, LU – Land use, FD – Fossil depletion.

The environmental impacts of 1 kg of packaged buffalo mozzarella cheese are reported in Table 5 and contributions of investigated stages along the supplied chain are shown in Fig. 2. The emissions related to CC was 12.03 ± 1.36 kg CO_2 eq., the activity for primary production accounted for about 90%, whereas energy and transports were the main contributors at cheese making plant and for the consumption, respectively. TA accounted for 102.20 ± 15.83 g SO_2 eq. and for about 95% referred to milk production and a lesser extent to cheese manufacturing and consumption. FE was 0.62 ± 0.12 g P eq., for about 70% was due to the cropping activity within farm gate, transports and cleaners were the main source of downstream processes after the farm gate. ME and OD were totally attributable to feed production. The contribution of milk production accounted for about 60% both for EC and HT, whereas consumers' habits and cheese making processes were the second hot spots, respectively. LU was associated with soymeal used to feed animals and FD was associated mainly with farm activities (60%). The remaining quota was equally shared between cheese making and home consumption.

3.1. Uncertainty analysis

The LCA results were employed to carry out an uncertainty analysis through 1000 Monte Carlo simulations and by using SimaPro v.9.1.1 software. To determine the qualitative uncertainty of input data was previously applied the Ecoinvent pedigree matrix approach [24], as indicated above on paragraph 2.5.

The Monte Carlo analysis combined the influence of all input uncertainty to calculate an uncertainty range of LCA results. It takes a random value from the uncertainty distribution of each input data and calculates and stores the LCA results for the sampled values. This procedure was run for 1000 simulations to form the uncertainty distribution of the final outcomes. Monte Carlo's results are reported in Table 6.

4. Discussion

The LCA is recognised worldwide as environmental impacts assessment methodology and has been already used in different studies to assess the environmental consequences of dairy buffalo farming at different stages of the supplied chain [12–14,17]. The LCA is a multicriteria environmental indicators that provides a broad view of the

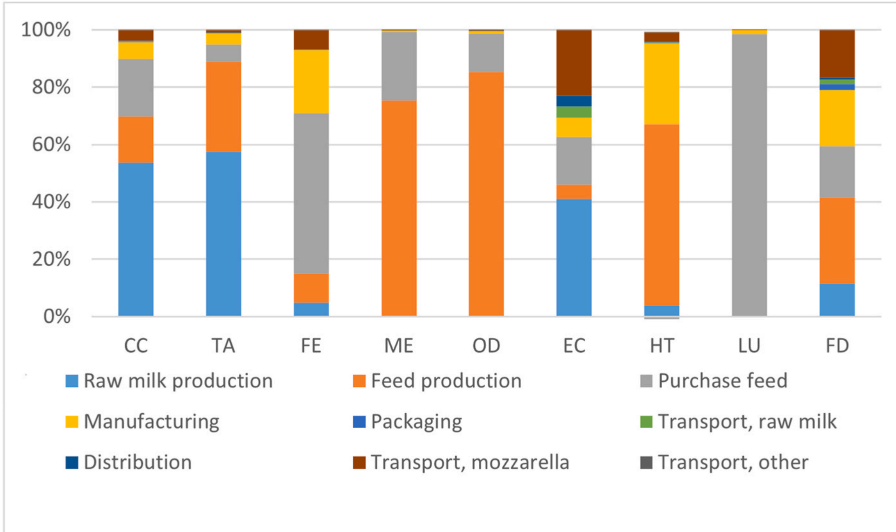

Fig. 2. Contribution analysis of the main pollution sources of the cradle-to-grave LCA of buffalo mozzarella cheese supply chain.
CC – Climate change, TA – Terrestrial acidification, FE – Freshwater eutrophication, ME – Marine eutrophication, OD – Ozone depletion, EC – Ecotoxicity, HT – Human toxicity, LU – Land use, FD – Fossil depletion.

Table 6
Uncertainty analysis using 1000 Monte Carlo simulation of 1 kg of buffalo mozzarella cheese from cradle-to-grave.

Impact categories	Unit	Mean	CV, %	95% CI	
CC	kg CO_2 eq.	12.03	6.06	10.97	13.83
TA	kg SO_2 eq.	1.02E-01	5.51	9.51E-02	1.18E-01
FE	kg P eq.	6.22E-04	25.37	3.70E-04	9.31E-04
ME	kg N eq.	9.45E-03	8.52	8.07E-03	1.13E-02
OD	kg CFC11 eq.	6.45E-05	7.37	6.90E-05	9.19E-05
EC	kg 1.4-DCB eq.	8.17	97.29	6.08	10.36
HT	kg 1.4-DCB eq.	4.64	248.57	1.58	7.79
LU	m^2a crop eq.	3.25	16.05	2.30	4.31
FD	kg oil eq.	8.83E-01	11.53	6.19E-01	9.68E-01

CV= Coefficient of variation; CI= Confidence interval.
CC – Climate change, TA – Terrestrial acidification, FE – Freshwater eutrophication, ME – Marine eutrophication, OD – Ozone depletion, EC – Ecotoxicity, HT – Human toxicity, LU – Land use, FD – Fossil depletion.

environmental pressure along life cycle of product and, in the present study, allowed to evaluate several environmental impacts related to the buffalo mozzarella cheese produced and consumed in central Italy.

According to published literature, CC was mostly affected by on-farm emissions due to animal and manure management. Enteric fermentation and manure storage impacted about 56% of CC and were responsible for methane and nitrous oxide emissions, which had a relevant weight on Global Warming Potential (GWP). The difference of GHG emissions across the investigated farms can be in part explained by diet quality and herd composition. Forage was the main ingredient in the diet with a variability among farms related to the number of lactating buffaloes and fattening beef that were fed with higher proportion of concentrate. The model developed to calculate the Y_m, a factor needed to estimate the amount of gross energy of the diet converted into methane, weights the digestibility of diet in a way that high content of forage is correlated with higher values of Y_m and higher enteric methane emissions [36]. The role of forage-to-concentrate ratio on enteric CH_4 was investigated in Holstein cows and the authors reported that the increasing of forage-to-concentrate ratio from 47:53 to 68:32 raised CH_4 from 538 to 648 g/cow/day [37]. The estimation of Y_m starting from forage-to-concentrate ratio allows to account diet quality which may improve the IPCC approach from tier 2 to 3. The forage-to-concentrate ratio of lactating buffaloes of our investigated farms ranged from 58:42 to 81:19 and the calculated values of Y_m were 6.21 and 7.16 that results in the estimated enteric methane emissions of 374 and 431 g/day/head, respectively. This approach increases the sensitivity of methane estimation, which represents the major source of GHG in ruminant systems [38], and allows to capture the quality of the diet as well as the effect of changes in feeding management.

Nutritional strategies can reduce methane from enteric fermentation, such as adding protected lipids on animals' diet [39]. This practice was applied on 35% of lactating buffalos considering the overall farms and the average reduction of CH_4 emissions was about 3.4%.

The model accounted the farms specific manure management such as slurry separation and anaerobic digestion that are conflicting to CC [40]. The model has evaluated the impact of both practices at reducing and increasing CH_4 and N_2O, respectively. The model also considered the increase of NH_3 generated by these practices [30]. Manure management practices may generate trade-offs regarding gasses emitted within the same impact category or between categories. Therefore, it is important to consider the "pollution swapping" effect when evaluating the effectiveness of manure management as mitigation strategy [38].

Manure storage and nitrogen fertilization (organic and inorganic) were the main drivers for ammonia emissions that was the major contributor (89%) to TA. The variation of TA among farms reflects differences in the manure management and fertilisers application. As reported above, the manure treatments applied in some of the investigated farms positively reduced GHG but, on the other hand, increased ammonia. However, best practices for manure storage to reduce NH_3 are available and they are aimed at decreasing the airflow across the surface by installing solid or floating covers, by allowing the formation of a surface crust, or by increasing the depth of stores to reduce the ratio of surface area to volume of the stores [41]. The implementation of these solutions in the investigated farms would contribute to reduce the TA.

An indirect method to mitigate impacts is through the improvement of production efficiency. This may be achieved by reducing the unproductive life of growing animals [14]. In addition to this, it should be considered that the dairy buffalo system is expanding and farms breed a high number of heifers compared to the long productive life of buffalo. Pirlo et colleagues (2014) pointed out that the heifers bred exceeding what needed to cover the culling rate can be considered as live weight leaving the farm and, in this case, the impact intensity of milk may be reduced. In the present study, we excluded one farm because of the high number of heifers, whereas heifers were compliant with declared culling rate for the nine enrolled farms. In light of this, we had not extra heifers to consider as live weight leaving the farm in the allocation procedure.

As reported by Sabia et al. [14] fertilizer application on field had a relevant impact on TA and ME categories. Acidification and eutrophication processes are affected by crops fertilization because of the nitrate-phosphate leaching and the ammonia volatilization related to this practice.

The techniques for the application of fertilisers may have a role at reducing NH_3 emission, in fact among farms we considered the mitigation impact of those techniques which were based on soil incorporation of organic and inorganic fertilisers [42].

Precision feeding is a recent concept of dietary management aimed to provide exactly what the animal needs to growth or produce and helps to reduce the release of mineral pollutants into manure and consequently into the environment. Therefore, this approach could be considered as valid indirect dietary mitigation strategy of TA and ME. The use of rumen protected amino acids instead of protein overfeeding comply with the concept of precision feeding that may positively reduce both ammonia volatilization and waste streams [43].

To account for ME, nitrogen was assumed as the limiting nutrient in the marine ecosystem [44] due to the high transfer of dissolved inorganic nitrogen from the soil and freshwater bodies to marine water.

Corn silage was the main on-farm crop in our dairy buffalo system and the high rate of mineral (0.2–0.3 ton/ha of mineral fertilization) and organic fertilisation contributed to ammonia emissions and nitrate leaching. The high supply of inorganic fertilizer on corn silage production was highlighted also by Bartl et al. [45], where emerged that nitrate and ammonia were the dominant sources of eutrophication processes.

On-farm feed production had a higher contribution also for the OD category because of N_2O emission which represents the most impactive ozone-depleting substance in this study. Lawrence et al. [46] reported a significant correlation between N_2O emissions and hydrological soil characteristics that can regulate rates of microbial nitrification and denitrification processes. Although N_2O emissions were produced as well from manure, in the present study the main share came from cropped soil meant as tillage and mineral fertilization. Farmers may contrast the contribution to this impact by reducing the quantity of N applied to the crops and preferring dry season for spreading.

In agreement with Pirlo et al. [12], purchased feed had a positive relationship with FE and had the main contribution in this impact category. Soybean and corn meal were used as main concentrate feed by most farms and were referred to extra-farm production.

The FE was closely related to nitrate and phosphate leaching, therefore field fertilisation connected to off-farm feeds affected nutrient levels on soil and increased P concentrations in fresh water. Also cheese making had a relevant effect on FE due to the substances realised into the environment related to cleaners' usage.

The production of concentrates for buffalos' diet was also the main source for LU category, which depended on the usage of arable and irrigated lands [14]. Cultivation of soybean had the highest impact on LU with a contribution of 59%. The relevant effect of this impact category was related to the increasing production of soybean which was expanded through cropland extensification into natural ecosystems [47].

The difference in buffalo's diet composition, especially in consideration to the different use of soybean meal, can explain the high variability of LU among investigated farms.

Mediterranean dairy buffalo have the highest milk yield compared with other buffaloes worldwide [48], and for this reason farmers need to sustain the high buffaloes' milk production with elevated energy and protein levels, as observed by Borghese [49]. Therefore, improving feeding efficiency and reducing the dependence of soybean meal with other protein sources can reduce the impact of LU and enhance environmental benefits. Nowadays, several studies have improved the knowledge about alternative protein sources in ruminants' diet. Halmemies-Beauchet-Filleau et al. [50] explored different options of low-impact grain legume protein and found out that fava beans (*Vicia faba*), peas (*Pisum sativum*) and lupins (*Lupinus* sp.) are comparable to soybean meal. In addition, these legume crops have the great potential to be sustainable cultivation in temperate zones and can permit to avoid the purchase of soy-based supplements.

Post-farm gate operations had the largest contribution to the EC category, accounting more than 40%. According to Eide [51] and Ledgard et al. [52], chemical compounds, heavy metals, and fossil fuels used for electricity production and transports are the main driver of EC. In our study this category was principally determined by transports throughout the supply chain, mainly referred to transports of purchased feed at farm and the transports of mozzarella from market to home. Additionally, EC was the only impact category where the HDPE packaging of mozzarella cheese shared a considerable contribution, this was due to the pollutants emitted for the production of plastic materials. The choise of packaging type for dairy products is crucial to preserve cheese quality and a long shelf life [53]. Several studies were aimed to improve food packaging within low environmental performance and the best solutions proposed were the use of lightweight materials or biomaterials and the addition of recycling technologies [19].

Milk and mozzarella were distributed through refrigerated lorries that resulted in high consumption of fossil fuel. As well as transports for distribution, the interviewed consumers have the habit to drive to cheese retailers with petrol or diesel cars that use fossil fuel. To mitigate the EC category can be useful to prefer electric or hybrid vehicles for the distribution of buffalo mozzarella cheese. Consumers can reduce emissions by using low-impact transports like electric vehicles or collective delivery systems which put together transports of different orders. In addition, can be useful to optimize the shopping trip including the purchase of more products [29].

The HT impact is associated with the risk increase of cancer or noncancer disease incidence, which was correlated to the environmental persistence, exposure, and toxicity of a compound throughout the human food chain [35]. In HT category the feed production was the main driver, especially due to diesel consumption for soil tillage. In addition, this impact category presented was also affected by diesel combustion to hot water at the cheese making plants.

Similarly, in the FD category flows of electricity and fossil fuel were relevant. This impact category esteemed the damage to fossil resource scarcity by determining the costs involved in fossil resource extraction and the economic burden that this current practice puts on future situations [54]. Impactive activities connected to diesel consumption were the same as HT, whereas the effects of energy usage linked with electricity were mainly due to the milking operations at farm and cheese making at dairy plant.

Since all investigated farms and dairies were supported by fossil resources, the effort to reduce energy consumption throughout the supply chain can lead to reduce FD impact, as reported by Kim et al. [18], in this sense an important advantage can be achieved by renewable energy use.

5. Conclusions

Through the LCA methodology it was possible to assess for the first time the environmental impacts of buffalo mozzarella cheese supply chain of central Italy, from milk production up to mozzarella consumption.

Farm activities give a large contribution to the majority of the investigated environmental impact categories. However, mitigation of these environmental impacts may be achieved through several strategies such as low-emitting practices of manure management and fertilizer application. Additionally, lipids supplementation and the use of alternative protein sources in animal diets may contribute to the reduction of enteric methane and land use change. Since energy usage is the primary contributor in cheese making and consumption stages, mitigation strategies based on the introduction of renewable energy sources and the improvement of energy efficiency of machineries with low-consuming technologies may contribute to improve the sustainability of supply chain.

The use of low-emitting vehicles and collective delivery systems may be valid options to reduce the impacts associated with distribution and consumption stages.

Although several options are available to mitigate environmental burdens associated to the life cycle of buffalo mozzarella cheese, in their implementation it is important to consider the "pollution swapping" effect that may limit the benefits of the mitigating strategy. This may occur when an action reduces an impact and, at the same time, it contributes to increase another one.

These results of the LCA analysis can provide more awareness to stakeholders and promote sustainable practices to reduce environmental impacts alongside the supply chain of buffalo mozzarella cheese.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jafr.2023.100871.

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